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GC-MS/MS profile of Acetone Fraction of Clematis zeylanica (L.)Pori. Leaves

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Abstract

Many of the chemicals used in pharmaceuticals, biochemical, fragrances, food colouring, and flavour, particularly in India, come from plants. Plant extracts has secondary metabolites, which are the wellsprings of numerous phytochemicals. Ethno-medicinal plant Clematis zeylanica is belongs to the family Ranunculaceae and it is utilized in the Indian Ayurveda System of Medication. The GC-MS/MS strategy is used for the examination of the acetone extracted sample; since it is a thrilling apparatus for explain the grouping of specific dynamic standards in medicinal plant utilized in the medication, drug and food enterprises. 124 peaks were observed in the GC-MS/MS chromatogram, indicating the presence of 124 bioactive compounds. This study uncovered the presence of different mixtures first time in quite a while like 11,13-Dihydroxy-tetradec-5-ynoic acid methyl ester (4.78%), N-(O-Nitrophenylthio)- l-leucine (4.73%), Parthenolide (4.25%), 5,7-Dodecadiyn-1,12-diol (3.72%), R-Limonene (3.19%), Benzeneacetaldehyde, .alpha.- (hydroxyimino)- , oxime (2.99%), Paromomycin (2.98%) and so on., The goal of this study was to decide the bioactive mixtures present in acetone leaf fraction of Clematis zeylainca (L.). Pori. The GC-MS/MS study revealed 124 bioactive compounds, which are useful to humankind to cure diseases.

Keywords: Clematis zeylanica leaves, Acetone extract, GC-MS/MS analysis, Parthenolide.

Introduction

BACKGROUND:

The worldwide capability of medicinal plants is significantly established in the way that many individuals, around 70-90%, depend on natural meds as their essential type of medical care. Medicinal plants have generally been huge wellsprings of bioactive substances that have been used to cure human sicknesses (WHO, 2002; Robinson and other, 2011; Rencken et al., 2015). A World Health Organization (WHO) master bunch characterized conventional medication as the sum of all information and practices — regardless of whether logical — utilized in the conclusion, counteraction, and end of physical, mental, or social lopsidedness and exclusively depending on direct insight and perception passed down orally or recorded as a hard copy from one generation to another (WHO, 1976). Natural medications are more secure than synthesized meds on the grounds that bio-constituents in plant extracts target the metabolic route. The majority of medicinal plants are one-of-a-kind in their capacity to treat a wide range of human ailments due to the participation of numerous important bio-constituents that are present in various plant sections.

The extraction and identification of various bioactive parts from a few medicinal plants has brought about the delivery of profoundly dynamic specialty medications. Utilizing chromatographic and spectrometric strategies to screen medicinal plants gives information on compound and pharmacological action, working with in the choice of naturally dynamic plants (Nagaraju et al., 2023). The ability of bioactive phytocompounds derived from various herbal plants to combat pathogens that cause diseases in humans and animals is well-known (Dewick, 2009; Tom et. al., 2018). Plants and trees serve an imperative job in medication, notwithstanding food and sustenance. Numerous dangerous illnesses have as of late been cured by making pastes from different plant parts like roots, barks, seeds, and leaves. The worldwide utilization of medicinal plants is expanding because of the far reaching utilization of conventional medication and the developing interest in herbal treatment (Idu, 2009).

Remarkably, such medicinal plant capacity property have attracted specialists to involve their lead synthetics for fostering the blend of current drugs. Over 25% of drug prescriptions accessible available today are gotten from medicinal plants (Gurib, 2006; Ha et al., 2018). The identification of bioactive components from medicinal plants that are commonly used locally and traditionally is a crucial step in drug discovery from medicinal plants. Since, Plants have given various essential necessities for humankind, including life-saving therapeutic mixtures (Akinyemi et al., 2018).

Clematis zeylanica is a types of blooming plant in the family Ranunculaceae. It is regularly known as Sri Lanka clematis and is local to the Indian subcontinent, specifically found in nations like India, Sri Lanka and Myanmar. Clematis zeylanica is a climbing plant that produces appealing white or cream shaded blossoms with a charming scent. Clematis zeylanica (L.) Pori Syn: Naravelia zeylanica (Linn) DC) has been utilized in the treatment of pitta, helminthiasis, dermatopathy, uncleanliness, rheumatalgia, odontalgia, colic aggravation, wounds, and ulcers in the Indian arrangement of medication Ayurveda (Raja Naika et al., 2008).

As per Ethnomedical Assessments, the flying bits of Naravelia zeylanica have generally been utilized in vitiated vata, pitta, irritation, and skin issues. At the point when the leaves are squashed, they discharge major fragrance that is breathed to treat colds, cerebral pains, and headaches. While root and stem paste is utilized externally to treat psoriasis, itches and skin sensitivities. It is used as a medication source in Kerala for intestinal worms, skin disease, sickness and toothache (Barlanka et al., 2013).

Conventional medication professionals in Karnataka are treating psoriasis and dermatitis with the leaf and stem juices. The root and stem have a strong penetrating scent and are utilized to treat malarial fever and cerebral pains. To treat wounds and worm infections, whole plant paste is applied externally to the affected area for two to three days (Mohan et al., 2008). In case of rhinitis, the dried stem is powdered and tied in a clean cloth bag and the smell from the cloth bag is delicately breathed in (Arun vijayan et al., 2007). Bubbling leaves in water and washing in it are utilized to reduce rheumatism (Suresh et al., 2011).

Unquestionably, the GC-MS/MS investigation of acetone extract of leaves of Clematis zeylanica is a significant scientific undertaking. This procedure gives a thorough understanding of the plant's chemical makeup. Permitting specialists to distinguish and evaluate different mixtures present inside it. This study's findings contribute to a better understanding of Clematis zeylanica plant complex molecular profile and its potential therapeutic applications and effects.

Methods:

2.1. Plant Collection:

The leaves of Clematis zeylanica L. (Pori) found in the Sanamavu Forest in the Krishnagiri district of Tamil Nadu, India. Were gathered at Latitude 12066’61”N and Longitude 77091’54”E. It is situated 9 kilometers southwest of Kelamangalam town and 8 kilometers southeast of the village of Samanapalli.

2.2. Identification of Plants:

The phytotherapeutic plant Clematis zeylanica L. (Pori.) was authenticated by Dr. M.U. SHARIEF, Scientist ‘F’ & Head, Botanical Survey of India, Tamil Nadu Agricultural University (TNAU) Campus, Coimbatore. The voucher number is BSI/SRC/5/23/2023/Tech.-749.

Systematic Position:	Clematis zeylanica (L.) Pori.	
Division	:	Tracheophyta
Class	:	Magnoliopsida
Order	:	Ranunculales
Family	:	Ranunculaceae
Genus	:	Clematis
Species	:	C. zeylanica (L.) Pori.

a b c

Fig. 1. Clematis zeylanica L. (Pori.) plant a. Habit b. Young twig c. Dried leaves powder.

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2.3. Leaves Extraction:

The gathered C. zeylanica leaves were cleaned with fresh running water and distilled water. Then, air dried at room temperature in the shade, powdered in electric blender and stored in a sealed container. The dried powdered substance is blended in with the acetone solvent (10mg/50ml). The blend was stored at room temperature for 72 hours with constant shaking. Using Whatman No. 41 filter paper, the extract and marc (the moist, solid substance) were separated. The extraction process was repeated three times. In preparation for GC-MS/MS analysis, the resulting extract was refrigerated.

2.4. Gas Chromatogram– Tandem Mass Spectroscopy (GC–MS/MS) analysis:

Clematis zeylanica L. (Pori.) leaf extracts were investigated utilizing an Agilent 7890B Gas Chromatography system equipped with a 7000C Mass Spectrometer Triple Quad (Agilent Innovations Inc. Sant Clara, CA, USA). The carrier gas was helium, which had a purity of 99.999%. The front inlet purge flow rate was 3 ml/min and the constant gas flow rate was 1.2 ml/min through the column. An Agilent HP-5ms UI column (30 m x 250 m x 0.25 m - Agilent Technologies Inc., Santa Clara, CA, USA) was used to separate volatile organic compounds (VOCs). The oven temperature program started at 1300 degrees Celsius for one minute, then ramped up to 2700 degrees Celsius at a rate of 100 degrees Celsius per minute, was held for 12 minutes, and the run length was set to 27 minutes. The Mass Selective Detector (MSD) transfer line, the Ion source and the Quadrupole mass detectors were all subjected to temperatures of 2900°C, 3000°C, and 1500°C, respectively. Mass Spectra in Electron Effect Ionizations (ME_EI) mode were distinguished at ionization energy of 70eV. The mass spectrometer's scanning mode was utilized to procure information from m/z 40 to 500. The solvent delayed for 3.2 minutes.

Result and disscussion:

The foundation of various phytochemical constituents found in medicinal plants can be identified by GC-MS/MS spectra analysis. The current work used GC-MS/MS analysis to look for phytochemical components in acetone extracts of Clematis zeylanica L. (Pori) leaves. Along with the retention time, Confirmation of Acceptance for Studies (CAS), molecular formal and peak of these extracts, numerous types of phytochemicals were found. A list of the bioactive compounds obtained from Clematis zeylanica was shown in Table 1. The GC-MS/MS analysis of the Clematis zeylanica acetone extract (CZAE) of leaves revealed that it had a greater quantity of higher peak chemicals [Figure.2].

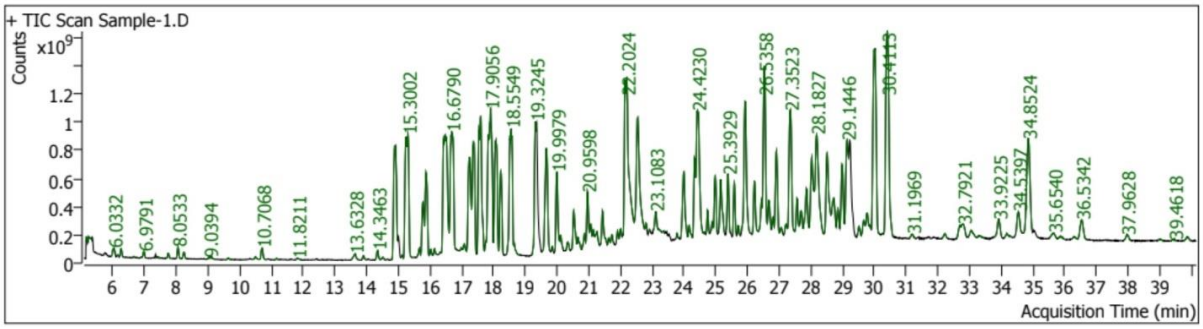

Fig. 2: GC-MS/MS chromatogram of acetone fraction of Clematis zeylanica L. leaves.

The GC-MS chromatogram of acetone leaves extract depicted different peaks resulting for the presence of 124 compounds. Based on abundance, the top compound identified was 11,13-Dihydroxy-tetradec-5-ynoic acid, methyl ester (4.78%) followed by N-(O-Nitrophenylthio)-l-leucine (4.73%), Parthenolide (4.25%), 5,7-Dodecadiyn-1,12-diol (3.72%), R-Limonene (3.19%), Benzeneacetaldehyde, .alpha.- (hydroxyimino)-, oxime (2.99%), Paromomycin (2.98%), 4-(2,5-Dihydro-3-methoxyphenyl)butylamine(2.91%), Nitrosothymol (2.76%), Benzeneethanol, .alpha.,.alpha.- dimethyl-, acetate(2.61%), 3-Acetamido-2-[2-methyl-1-propenyl]phenol (2.57%), (-)-Isolongifolol, acetate(2.34%) Parthenolide (2.38%) Brefeldin A, 2Ac derivative (2.22%), Panaxydol (1.92%), 3-(2-Aminopropyl)phenol (1.81%),1H-3a,7-Methanoazulene, octahydro-3,8,8-trimethyl-6- methylene-, [3R-(3.alpha.,3a.beta.,7.beta.,8a.alpha.)](1.74%) 1H-Cycloprop[e]azulene, 1a,2,3,5,6,7,7a,7b-octahydro-1,1,4,7-tetramethyl-, [1aR-(1a.alpha.,7.alpha.,7a.beta.,7b.alpha.)]- (1.67%) Ethyl 2-benzamido-3,3,3-trifluoro-2-(3-methoxybenzylamino) propionate(1.62%), (8R,8aS)-8,8a-Dimethyl-2-(propan-2-ylidene)-1,2,3,7,8,8a- hexahydronaphthalene (1.44%) 1-(4-Chloro-phenyl)-3-(2-pyridin-2- yl-ethylamino)-pyrrolidine-2,5-dione(1.40%) Longifolene (1.25%) Cyclohexanol, 3-(acetyloxymethyl)-2,2,4-trimethyl- (1.22%) 11- Oxatetracyclo [5.3.2.0(2,7).0(2,8)]do decan-9-one (1.21%) 7-Oxabicyclo[4.1.0]heptane, 1- methyl-4-(2-methyloxiranyl)- (1.13%) Benzene, 1-methyl-4-(1- methylethyl)-2-nitro- (1.12%), etc.,

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Table 1: Compounds from the acetone leaves extract of Clematis zeylanica by GC-MS/MS.

S. No.	RT	Compound Name	Peak Area	Area %	CAS#	Formula
1	5.1514	2-Oxopentanedioic acid	372980483.2	0.17	328-50-7	$C_5H_6O_5$
2	5.2476	9-Octadecenamide, (Z)-	849949937.9	0.39	301-02-0	$C_{18}H_{35}NO$
3	6.0332	Pentane, 2-methyl-	258160649.5	0.12	107-83-5	C_6H_{14}
4	6.1454	Methadone	24571190.0	0.01	76-99-3	$C_{21}H_{27}NO$
5	6.2576	1,3 Propanediol, 2	182397465.6	0.08	77-99-6	$C_6H_{14}O_3$
6	6.6585	Morpholine,4-[5-(2-furfurylthio)-2- nitrophenyl]-	23059665.3	0.01	1000272-46-5	$C_{15}H_{16}N_2O_4S$
7	6.9791	Cyclopropane,1-ethyl-1-methyl-	169715329.2	0.08	53778-43-1	C_6H_{12}
8	7.3960	3-(2 Benzoxazolyl) benzenamine, N-trimethylsilyl-	20358863.2	0.01	1000471-59-4	$C_{16}H_{18}N_2OSi$
9	7.7487	Cyclohexane	118079673.4	0.05	110-82-7	C_6H_{12}
10	8.0533	Butanal, 3-methyl-	247307911.1	0.11	590-86-3	$C_5H_{10}O$
11	8.2377	Butanal, 2-methyl-	160703380.0	0.07	96-17-3	$C_5H_{10}O$
12	9.0394	8,11,14-Eicosatrienoic acid, methylester, (Z,Z,Z)-	47616195.0	0.02	21061-10-9	$C_{21}H_{36}O_2$
13	9.1115	2,3-dimethylfuran	58447319.2	0.03	1000458-49-9	C_6H_8O
14	9.6246	3,4-Dihydroxybenzaldehyde, bis(trimethylsilyl) ether	34767339.0	0.02	10586-13-7	$C_{13}H_{22}O_3Si_2$
15	10.4984	Z,Z,Z-4,6,9-Nonadecatriene	94884188.9	0.04	1000131-11-7	$C_{19}H_{34}$
16	10.7068	1,5-Heptadien-3-yne	349875657.6	0.16	3511-27-1	C_7H_8
17	11.1637	9,12-Hexadecadienoic acid, methyl ester	33063904.4	0.02	2462-80-8	$C_{17}H_{30}O_2$
18	11.8211	Dodecane, 1,2-dibromo-	55986753.6	0.03	55334-42-4	$C_{12}H_{24}Br_2$
19	13.6328	(+)-4-Carene	244598609.8	0.11	29050-33-7	$C_{10}H_{16}$
20	13.8973	2-Pentanone, 4-hydroxy-4-methyl-	107003981.5	0.05	123-42-2	$C_6H_{12}O_2$
21	14.3463	Bicyclo[2.2.1]hept-2-ene, 1,7,7-trimethyl-	251910358.1	0.12	464-17-5	$C_{10}H_{16}$
22	14.5066	2-Heptanone	67877569.2	0.03	110-43-0	$C_7H_{14}O$
23	14.8834	5,7-Dodecadiyn-1,12-diol	5156299549.4	2.39	74602-32-7	$C_{12}H_{18}O_2$
24	15.1239	13-Heptadecyn-1-ol	113483943.9	0.05	56554-77-9	$C_{17}H_{32}O$
25	15.2441	4-(2,5-Dihydro-3-methoxyphenyl)butylamine	3632678983.1	1.68	77515-67-4	$C_{11}H_{19}NO$
26	15.3002	Ethyl2-benzamido-3,3,3-trifluoro-2-(3-methoxy benzylamino) propionate	3489730888.4	1.62	1000224-75-5	$C_{20}H_{21}F_3N_2O_4$
27	15.6610	.alpha.,d-Gala-octonic phenylhydrazide	251024990.9	0.12	110439-22-0	$C_{14}H_{22}N_2O_8$
28	15.7732	7-Methylene-9-oxa-bicyclo[3.3.1]non-2-ene	1787335815.4	0.83	1000193-85-5	$C_9H_{12}O$
29	15.8694	1-(4-Chloro-phenyl)-3-(2-pyridin-2- yl-ethylamino)-pyrrolidine-2,5-dione	3013007289.6	1.40	1000301-18-1	$C_{17}H_{16}ClN_3O_2$
30	16.0217	Cinnamyl carbanilate	120467052.9	0.06	25076-44-2	$C_{16}H_{15}NO_2$
31	16.1419	2,4,6-Octatriene, 3,4-dimethyl-	130858394.3	0.06	57396-75-5	$C_{10}H_{16}$
32	16.4867	5,7-Dodecadiyn-1,12-diol	8031268980.8	3.72	74602-32-7	$C_{12}H_{18}O_2$
33	16.6790	4-(2,5-Dihydro-3-methoxyphenyl)butylamine	6282062039.7	2.91	77515-67-4	$C_{11}H_{19}NO$
34	16.9837	6-Hydroxy-10a-methyl-4,7-dimethylidene-2,8-dioxododecahydrooxireno[8,9] cyclodeca [1,2-b]furan-5-yl (2Z)-2- methylbut-2-enoate	94702812.4	0.04	1798297-60-5	$C_{20}H_{24}O_7$
35	17.0799	2,4,6-Octatriene, 3,4-dimethyl-	219664774.1	0.10	57396-75-5	$C_{10}H_{16}$
36	17.2402	3-(2-Aminopropyl)phenol	3904368305.5	1.81	1075-61-2	$C_9H_{13}NO$
37	17.3685	4-(2,5-Dihydro-3-methoxyphenyl)butylamine	4490579967.4	2.08	77515-67-4	$C_{11}H_{19}NO$

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38	17.5929	R-Limonene	6878614728.6	3.19	1000371-50-6	$C_{10}H_{16}O_3$
39	17.8575	4-(2,5-Dihydro-3-methoxyphenyl)butylamine	3394035450.7	1.57	77515-67-4	$C_{11}H_{19}NO$
40	17.9056	Benzeneethanol, .alpha.,.alpha.- dimethyl-, acetate	5627263398.1	2.61	151-05-3	$C_{12}H_{16}O_2$
41	18.0739	Paromomycin	4921969467.9	2.28	7542-37-2	$C_{23}H_{45}N_5O_{14}$
42	18.2262	Cyclohexanol, 3-(acetyloxymethyl)-2,2,4-trimethyl-	2643386591.5	1.22	1000162-14-5	$C_{12}H_{22}O_3$
43	18.5549	Paromomycin	6439227774.5	2.98	7542-37-2	$C_{23}H_{45}N_5O_{14}$
44	19.3245	5,7-Dodecadiyn-1,12-diol	2764011931.1	1.28	74602-32-7	$C_{12}H_{18}O_2$
45	19.6612	Panaxydol	4145009470.9	1.92	72800-72-7	$C_{17}H_{24}O_2$
46	19.8536	2-Hexanone, 3-[hydroxy(2-nitrophenyl)methyl]-	71914224.9	0.03	62238-21-5	$C_{13}H_{17}NO_4$
47	19.9979	7-Oxabicyclo[4.1.0]heptane, 1-methyl-4-(2-methyloxiranyl)-	2445632192.2	1.13	96-08-2	$C_{10}H_{16}O_2$
48	20.1101	Spiro[3.5]nona-5,7-dien-1-one,	448381444.8	0.21	91531-36-1	$C_{12}H_{16}O$
49		5,9,9-trimethyl-				
50	20.3426	Bicyclo[3.1.0]hexane-6-methanol, 2- hydroxy-1,4,4-trimethyl-	241031877.6	0.11	58795-41-8	$C_{10}H_{18}O_2$
51	20.5270	4-Isopropyl-1-methylcyclohex-2-enol	1342258928.7	0.62	619-62-5	$C_{10}H_{18}O$
52	20.6632	Benzene, 2-ethyl-1,4-dimethyl-	422237835.9	0.20	1758-88-9	$C_{10}H_{14}$
53	20.8636	6,7-Dimethyl-3,5,8,8a-tetrahydro-1H-2-benzopyran	458256602.7	0.21	110028-10-9	$C_{11}H_{16}O$
54	20.9598	Cyclohexanol, 3-(acetyloxymethyl)- 2,2,4-trimethyl-	1609202032.7	0.75	1000162-14-5	$C_{12}H_{22}O_3$
55	21.0641	Octadecanal, 2-bromo-	525916191.7	0.24	56599-95-2	$C_{18}H_{35}BrO$
56	21.1442	1-Cyclopentene-1-methanol, .alpha.,.alpha.,4,5-tetramethyl-, trans-	396201997.3	0.18	56666-69-4	$C_{10}H_{18}O$
57	21.2564	1-Methoxybicyclo[2,2,2]oct-5-en-2-yl methyl ketone	446042874.1	0.21	1000132-14-1	$C_{11}H_{16}O_2$
58	21.4328	4-Isopropyl-1-methylcyclohex-2-enol	1138827752.8	0.53	619-62-5	$C_{10}H_{18}O$
59	21.6332	2-Oxaspiro[5.5]undecane-3,5-dione, 1-(3,4-dimethoxyphenyl)-4,4-dimethyl-	129328991.0	0.06	1000277-11-3	$C_{20}H_{26}O_5$
60	21.7294	3-Isopropylidene-5-methyl-hex-4-en-2-one	240646631.9	0.11	64149-32-2	$C_{10}H_{16}O$
61	21.9058	Bicyclo[2.2.1]heptan-2-ol, 2,3,3- trimethyl-	195198117.1	0.09	465-31-6	$C_{10}H_{18}O$
62	22.0100	(+)-2-Bornanone	187076614.2	0.09	464-49-3	$C_{10}H_{16}O$
63	22.2024	N-(O-Nitrophenylthio)-l-leucine	10203684413.2	4.73	7685-67-8	$C_{12}H_{16}N_2O_4S$
64	22.5471	Paromomycin	5911061083.3	2.74	7542-37-2	$C_{23}H_{45}N_5O_{14}$
65	22.8036	4,11-Dispiro(2'-cyclobutanone)tricyclo[6.2.2.0(2,7)] dodeca-5,9-diene, 1,3,3,5,12,12- hexamethyl-	172039800.6	0.08	1000191-30-1	$C_{24}H_{32}O_2$
66	22.9159	2,2,4-Trimethyl-3-(3,8,12,16-tetramethyl-heptadeca-3,7,11,15- tetraenyl)-cyclohexanol	68256470.5	0.03	1000194-01-4	$C_{30}H_{52}O$
67	23.1083	Ethanol, 2-(9,12-octadecadienyloxy)- , (Z,Z)-	759137702.8	0.35	17367-08-7	$C_{20}H_{38}O_2$
68	23.3728	3,9-Epoxypregn-16-en-20-one, 3-	25053123.1	0.01	1000194-70-2	$C_{28}H_{38}O_9$
69	23.7816	Cyclopropa[5,6]-A-nor-5.alpha.- androstane-3,7-dione, 3',6.beta.- dihydro-17.beta.-hydroxy-3',3'- dimethyl-, acetate	66642894.9	0.03	24634-23-9	$C_{23}H_{32}O_4$
70	23.7816	Cyclopropa[5,6]-A-nor-5.alpha.-	66642894.9	0.03	24634-23-9	$C_{23}H_{32}O_4$

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71	23.9981	11-Oxatetracyclo [5.3.2.0(2,7).0(2,8)] do decan-9-one	2607415420.5	1.21	1000186-25-4	$C_{11}H_{14}O_2$
72	24.1664	2-Undecanone	390223098.0	0.18	112-12-9	$C_{11}H_{22}O$
73	24.3348	Acetic acid, 1,7,7-trimethyl-bicyclo[2.2.1]hept-2-yl ester	1965089640.5	0.91	92618-89-8	$C_{12}H_{20}O_2$
74	24.4230	Benzeneacetaldehyde, .alpha.-(hydroxyimino)-, oxime	6456907643.3	2.99	4589-97-3	$C_8H_8N_2O_2$
75	24.7436	Carane, 4,5-epoxy-, trans	468739647.6	0.22	6909-20-2	$C_{10}H_{16}O$
76	24.8959	6-(1-Adamantylamino)-2,4,5-trichloronicotinonitrile	255980879.7	0.12	339165-88-7	$C_{16}H_{16}Cl_3N_3$
77	24.9841	2-Oxa-6-azatricyclo[3.3.1.1(3,7)]decane	1641666525.5	0.76	19557-29-0	$C_8H_{13}NO$
78	25.1605	1,2,4-Metheno-1H-indene, octahydro-1,7a-dimethyl-5-(1-methylethyl)-, [1S-(1.alpha.,2.alpha.,3a.beta.,4.alpha., 5.alpha.,7a.beta.,8S*)]-	1860966015.0	0.86	22469-52-9	$C_{15}H_{24}$
79	25.3929	Terpin diacetate, trans-	1549464133.9	0.72	1000159-39-5	$C_{14}H_{24}O_4$
80	25.5853	5-Hydroxy-2,3,3-trimethyl-2-(3- methyl-buta-1,3-dienyl)-cyclohexanone	1212929084.4	0.56	1000189-10-4	$C_{14}H_{22}O_2$
81	25.7136	(R)-1-Methyl-4-(6-methylhept-5-en-2-yl)cyclohexa-1,4-diene	188183292.3	0.09	28976-67-2	$C_{15}H_{24}$
82	25.9300	Parthenolide	5144017828.8	2.38	20554-84-1	$C_{15}H_{20}O_3$
83	26.2186	Cyclohexane, 1-ethenyl-1-methyl-2,4-bis(1-methylethenyl)-, [1S-(1.alpha.,2.beta.,4.beta.)]-	1520170407.3	0.70	515-13-9	$C_{15}H_{24}$
84	26.4431	cis-5,8,11,14,17-Eicosapentaenoic acid	804335478.2	0.37	10417-94-4	$C_{20}H_{30}O_2$
85	26.5358	Nitrosothymol	5955772127.8	2.76	2364-54-7	$C_{10}H_{13}NO_2$
86	26.6756	(+)-2-Carene, 2-isopropenyl-	1123277729.1	0.52	1000151-27-0	$C_{13}H_{20}$
87	26.8038	Phenol, 2-(1-methyl-2-buthenyl)-4- methoxy-	398190317.8	0.18	95414-66-7	$C_{12}H_{16}O_2$
88	26.9159	Longifolene	2707825398.2	1.25	475-20-7	$C_{15}H_{24}$
89	27.0684	Ylangene	144998658.0	0.07	14912-44-8	$C_{15}H_{24}$
90	27.2207	1H-3a,7-Methanoazulene, 2,3,4,7,8,8a-hexahydro-3,6,8,8- tetramethyl-, [3R-(3.alpha.,3a.beta.,7.beta.,8a.alpha.)]	249376252.2	0.12	469-61-4	$C_{15}H_{24}$
91	27.3523	(-)-Isolongifolol, acetate	5052995432.4	2.34	1000352-28-0	$C_{17}H_{28}O_2$
92	27.5654	(E)-2-((8R,8aS)-8,8a-Dimethyl-3,4,6,7,8,8a-hexahydronaphthalen-2(1H)-ylidene)propan-1-ol	1028589681.0	0.48	22387-74-2	$C_{15}H_{24}O$
93	27.6937	(3R,4R,5R)-3-[(11E)-Hexadeca-11,15-dien-9-ynyl]-4-hydroxy-5- methyloxolan-2-one, O-acetyl	767682478.1	0.36	1000494-29-7	$C2_3H_{34}O_4$
94	27.8620	Spiro[5.5]undec-2-ene, 3,7,7-trimethyl-11-methylene-, (-)-	1700642370.2	0.79	18431-82-8	$C_{15}H_{24}$
95	28.0304	(8R,8aS)-8,8a-Dimethyl-2-(propan-2-ylidene)-1,2,3,7,8,8a-hexahydronaphthalene	3099713249.5	1.44	27840-40-0	$C_{15}H_{22}$
96	28.1827	3-Acetamido-2-[2-methyl-1-propenyl]phenol	5554476904.4	2.57	31011-04-8	$C_{12}H_{15}NO_2$
97	28.5113	1H-Cycloprop[e]azulene, 1a,2,3,5,6,7,7a,7b-octahydro-1,1,4,7-tetramethyl-, [1aR-(1a.alpha.,7.alpha.,7a.beta.,7b.alpha.)]-	3601270497.1	1.67	21747-46-6	$C_{15}H_{24}$
98	28.7198	.alpha.-Muurolene	1830494331.1	0.85	10208-80-7	$C_{15}H_{24}$
99	28.8561	(4aS,9aR)-3,5,5,9-Tetramethyl-2,4a,5,6,7,9a-hexahydro-1H-benzo[7]annulene	795962682.6	0.37	53111-25-4	$C_{15}H_{24}$

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100	28.9843	Benzene, 1-methyl-4-(1-methylethyl)-2-nitro-	2424502075.7	1.12	943-15-7	$C_{10}H_{13}NO_2$
101	29.1446	1H-3a,7-Methanoazulene, octahydro-3,8,8-trimethyl-6-methylene-, [3R-(3.alpha.,3a.beta.,7.beta.,8a.alpha.)]	3756129518.1	1.74	546-28-1	$C_{15}H_{24}$
102	29.5214	Naphthalene, 1,2,4a,5,8,8a-(1.alpha.,4a.beta.,8a.alpha.)-(.+/-.)-	275820004.9	0.13	5951-61-1	$C_{15}H_{24}$
103	29.6657	4-Isopropyl-6-methyl-1-tetralone	500070851.0	0.23	57494-10-7	$C_{14}H_{18}O$
104	29.7780	1H-3a,7-Methanoazulene, octahydro-3,8,8-trimethyl-6-methylene-, [3R-(3.alpha.,3a.beta.,7.beta.,8a.alpha.)]	1071840203.1	0.50	546-28-1	$C_{15}H_{24}$
105	30.0184	Parthenolide	9174222848.6	4.25	20554-84-1	$C_{15}H_{20}O_3$
106	30.4113	11,13-Dihydroxy-tetradec-5-ynoic acid, methyl ester	10324676237.4	4.78	1010193-81-6	$C_{15}H_{26}O_4$
107	31.1969	7,10,13-Eicosatrienoic acid, methyl ester	164165250.0	0.08	30223-51-9	$C_{21}H_{36}O_2$
108	32.2310	(E)-2-((8R,8aS)-8,8a-Dimethyl-3,4,6,7,8,8a-hexahydronaphthalen-2(1H)-ylidene)propanal	205589414.1	0.10	137695-18-2	$C_{15}H_{22}O$
109	32.7921	(1R,2S,6S,7S,8S)-8-Isopropyl-1-methyl-3-methylenetricyclo[4.4.0.02,7]decane-rel-	1197307435.7	0.55	18252-44-3	$C_{15}H_{24}$
110	33.0487	Phenol, 2,6-dimethoxy-4-(2-propenyl)-	355780420.4	0.16	6627-88-9	$C_{11}H_{14}O_3$
111	33.3292	1H-Cyclopropa[3,4]benz[1,2-e]azulene-5,7b,9,9a-tetrol, 3-[(acetyloxy)methyl]-1a,1b,4,4a,5,7a,8,9-octahydro-1,1,6,8-tetramethyl-, 9,9a-diacetate, [1aR-(1a.alpha.,1b.beta.,4a.alpha.,5.beta.,7a.alpha.,7b.alpha.,8.alpha.,9.beta., 9a.alpha.)]-	100853025.5	0.05	77550-15-3	$C_{26}H_{36}O_8$
112	33.9225	.beta.-Asarone	821805986.9	0.38	5273-86-9	$C_{12}H_{16}O_3$
113	34.1790	Folic Acid	156899523.0	0.07	59-30-3	$C_{19}H_{19}N_7O_6$
114	34.5397	2,6-Dimethyl-10-methylene-12-oxatricyclo[7.3.1.0(1,6)]tridec-2-ene	879914041.3	0.41	1000191-85-5	$C_{15}H_{22}O$
115	34.8524	Brefeldin A, 2Ac derivative	4792444656.2	2.22	29129-37-1	$C_{20}H_{28}O_6$
116	35.6540	1H-Cyclopropa[3,4]benz[1,2-e]azulene-4a,5,7b,9,9a(1aH)-pentol, 3-[(acetyloxy)methyl]-1b,4,5,7a,8,9-hexahydro-1,1,6,8-tetramethyl-,5,9,9a-triacetate, [1aR-(1a.alpha.,1b.beta.,4a.beta.,5.beta.,7a.alpha.,7b.alpha.,8.alpha.,9.beta., 9a.alpha.)]-	317725950.9	0.15	77698-37-4	$C_{28}H_{38}O_{10}$
117	35.8785	5H-Cyclopropa[3,4]benz[1,2-e]azulen-5-one, 9,9a-bis(acetyloxy)-3-[(acetyloxy)methyl]-1,1a,1b,2,3,4,4a,7a,7b,8,9,9a-dodecahydro-2,3,4a,7b-tetrahydroxy-1,1,6,8-tetramethyl-,[1ar-(1a.alpha.,1b.beta.,2.alpha.,3.alpha.,4a.beta.,7a.alpha.,7b.alpha.,8.alpha.,9.beta.,9a.alpha.)]-	125565173.0	0.06	77646-82-3	$C_{26}H_{36}O_{11}$
118	36.2953	6-Hydroxy-10a-methyl-4,7-dimethylidene-3,8-dioxododecahydrooxireno[8,9]cyclod eca[1,2-b]furan-5-yl	103143587.6	0.05	1217869-57-2	$C_{20}H_{24}O_7$

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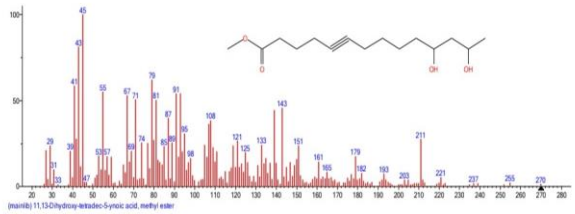
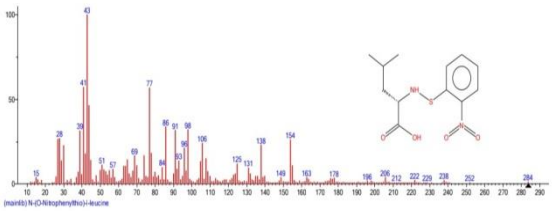
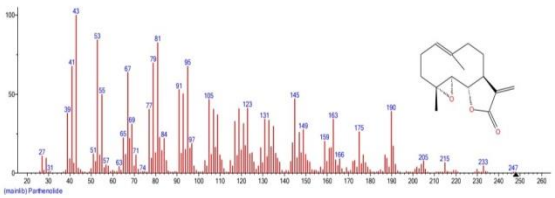
		(2Z)-2- methylbut-2-enoate				
119	36.5342	Bicyclo[10.1.0]trideca-4,8-diene-13- carboxamide, N-(3,4- dimethylphenyl)-	1023187977.1	0.47	322416-89-7	$C_{22}H_{29}NO$
120	37.9628	Tetradecanoic acid, ethyl ester	278330300.0	0.13	124-06-1	$C_{16}H_{32}O_2$
121	39.0049	9-Desoxo-9x-hydroxy-7-ketoingol 3,8,9,12-tetraacetate	76877390.4	0.04	1000203-03-0	$C_{28}H_{38}O_{10}$
122	39.4618	Pregn-16-en-20-one, 11,18-	78162441.8	0.04	30384-39-5	$C_{26}H_{36}O_7$
123		bis(acetyloxy)-3,9-epoxy-3-methoxy-, (3.alpha.,5.beta.,11.alpha.)-				
124	39.8787	2-Butenoic acid, 2-methyl-, 2-(acetyloxy)-1,1a,2,3,4,6,7,10,11,11a-decahydro-7,10-dihydroxy-1,1,3,6,9- pentamethyl-4a,7a-epoxy-5H-cyclopenta[a]cyclopropa[f]cyclounde cen-11-yl ester, [1aR-[1aR*,2R*,3S*,4aR*,6S*,7S*,7aS*,8 E,10R*,11R*(E),11aS*]]-	178187523.8	0.08	51906-13-9	$C_{27}H_{38}O_8$



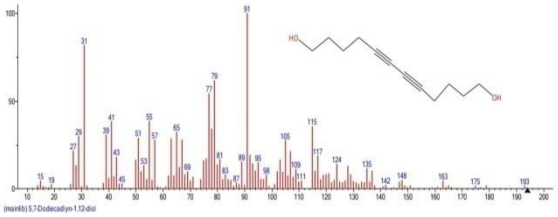
a). 11,13-Dihydroxy-tetradec-5-ynoic acid, methyl ester



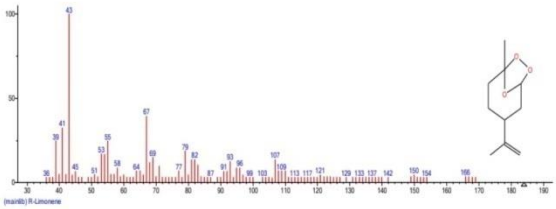
d). N-(O-Nitrophenylthio)-l-leucine



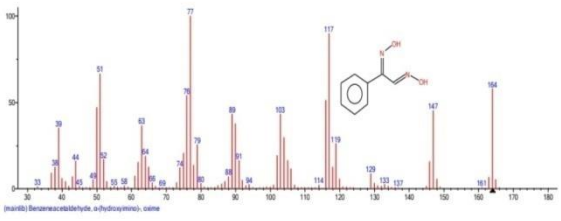
b). Parthenolide



e). 5,7-Dodecadiyn-1,12-diol



c). R-Limonene



f) Benzeneacetaldehyde, .alpha.- (hydroxyimino)- oxime

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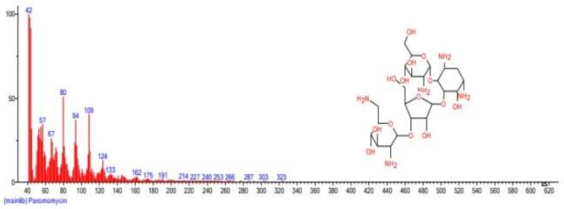
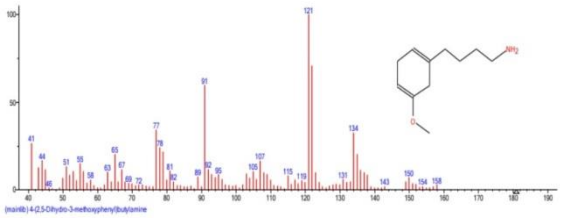
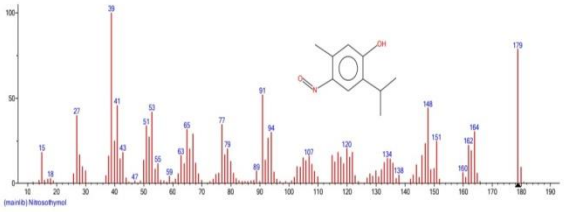


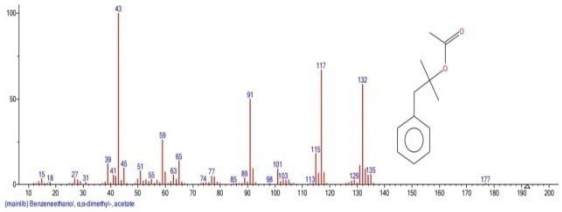
g). Paromomycin



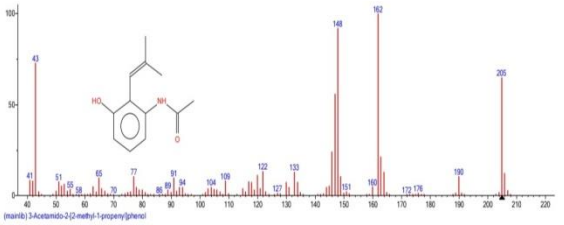
i). 4-(2,5-Dihydro-3-methoxyphenyl)butylamine



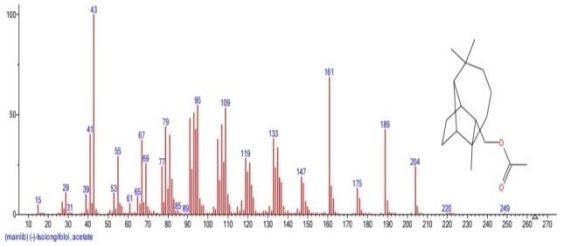
h). Nitrosothymol



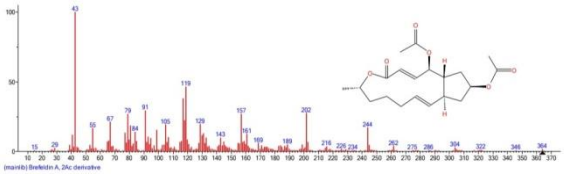
j). Benzeneethanol, .alpha.,.alpha.- dimethyl-, acetate



3-Acetamido-2-[2-methyl-1- propenyl]phenol



(-)-Isolongifolol, acetate



Brefeldin A, 2Ac derivative

Fig: 3. Identified major compounds from acetone extract of Clematis zeylanica L. Leaves.

4-(2,5-Dihydro-3- methoxyphenyl)butylamine - Antitumor, antispasmolytic, estrogenic, antiviral (Hussein et al. 2016) Paromomycin used as antimicrobial agents because it shows antimicrobial activity (Kamal, 2017). 5,7-Dodecadiyn-1,12-diol used as antifungal agent (Kamal, 2017). N-(O-Nitrophenylthio)-l-leucine was previously identified in Chenopodium album leaves (Javaid, 2023) but pharmaceutical activities unknown (Sabreen et al., 2017). 11,13-Dihydroxy-tetradec-5-ynoic acid, methyl ester was previously identified in Citrullus colocynthis and Caesalpinia decapetula (Thakur et al., 2017), but pharmaceutical activities are unknown.

A brand-new substance called parthenolide (PTL or PAR) has been widely used as a herbal medicine for various heath conditions. It was also derived from traditional Chinese medicine. Its indisputable involvement in tumours, migraine, arthritis, autoimmune disorders, and inflammatory diseases has been demonstrated in recent years (Pareek et al., 2011; Hall et al., 1980; Heinrich et al., 1998, Jiye Liu, 2023). It is a secondary metabolite of Compositae (Daisy) and is a completely natural sesquiterpene lactone (Freund, 2020). Feverfew (Tanacetum parthenium) was the source of its initial extraction (Suthar, 2011). PTL also has anti-inflammatory effect in respiratory diseases and antioxidant properties (Joanna et al. 2020).

Numerous studies on the medicinal properties of limonene have revealed its anti-inflammatory, antioxidant, antinociceptive, anticancer, antidiabetic, antihyperalgesic, antiviral, and gastroprotective properties, among other health-promoting activities (Vieira et al. 2018). Benzeneacetaldehyde, .alpha.- (hydroxyimino)-, oxime is phamacological activity unknown. Benzeneethanol, .Alpha.,.Alpha.-Dimethyl-, Acetate reported in the GC-MS analysis of Wedlia biflora leaves methanol extract (Arockia Sahayarah et al. 2015) but Pharmacological activity unknown. A polyacetylene chemical called panaxydol that was identified from Panax ginseng has anti-proliferative actions on cancer cells (Guo, 2009). 3-acetamido-2-(2-methyl-1-propenyl) phenol pharmacological activity is unknown. Brefeldin A ester derivatives (BFA) are potential anticancer agents (Anadu et al., 2006).

Table 2: Biological activity of specific phytochemicals identified in acetone extract of Leaves of C. zeylanica (L.)Pori.

S.No	Name of the compound	Biological activities
1.	11,13-Dihydroxy-tetradec-5-ynoic acid, methyl ester	Unknown
2.	N-(O-Nitrophenylthio)-l-leucine	Unknown
3.	Parthenolide	Tumours, migraine, arthritis, autoimmune disorders, inflammatory diseases respiratory diseases and antioxidant.
4.	5,7-Dodecadiyn-1,12-diol	Antifungal agent
5	R-Limonene	Anti-inflammatory, antioxidant, antinociceptive, anticancer, antidiabetic, antihyperalgesic, antiviral, and gastroprotective properties, among other health-promoting activities.
6.	Benzeneacetaldehyde, .alpha.-(hydroxyimino)-, oxime	Unknown
7.	Paromomycin	Antimicrobial agents
8.	4-(2,5-Dihydro-3-methoxyphenyl)butylamine	Antitumor, antispasmolytic, estrogenic, antiviral.
9.	Thymol	Antioxidant, anti-inflammatory, anti-microbial.
10.	Benzeneethanol, .alpha.,.alpha.-dimethyl-, acetate	Unknown
11.	3-Acetamido-2-[2-methyl-1-propenyl]phenol	Unknown
12.	Brefeldin A	Anticancer agent

Conclusion:

The GC-MS/MS study revealed that the Clematis zeylanica acetone extract (CZAE) of leaves contained 124 bioactive compounds. As a result, the plant is believed to be pharmaceutically relevant, and separation of particular bioactive components may be required to discover a novel medication. Additional research is required to extract specific active ingredient and to explain the exact system of activity in treating different ailments and foster novel medications.

Conflict of interest:

On behalf of all authors, the corresponding author states that there is no conflict of interest.

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