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GC-MS analysis of ethanol extract of aerial and underground parts of *Cyperus rotundus* L.

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Abstract
Cyperus rotundus L., frequently used in the traditional medicine for inflammatory disorders. The present study was carried to find out the possible bioactive components in the ethanol extract of aerial and underground parts of this plant by Gas Chromatography-Mass Spectrometry (GC-MS) analysis. The mass spectra of the extract found compounds that were matched with the National Institute of Standards and Technology (NIST) library database. The GC-MS study observed different peaks as per retention time and then those peaks were analyzed determining presence of 19 and 21 phytochemical compounds in aerial and underground parts respectively.

Keywords: *Cyperus rotundus*, aerial and underground parts, bioactive components, GC-MS analysis.

Introduction
Cyperus rotundus L. a member of the family Cyperaceae is found all over India. It is widely distributed and grown in many regions of the world, and in some regions, it is even regarded as a weed [5, 19]. The name Cyperus is derived from Cypeiros, the genus's old Greek name; rotundus, a Latin term meaning "round," alludes to the tuber [6, 10].
It is an erect, glabrous, grass like, perennial herb that can grow up to 7–40-cm height and proliferate widely by rhizomes and tubers [14]. The plant produces rosettes of leaves, scapes, and umbels above ground and rhizomes, tubers, basal bulbs, and fibrous roots underground [10].
C. rotundus is a medicinal herb used in folk medicine in several countries including India, Iran, China, Japan, Vietnam, etc. [12, 16, 26]. *C. rotundus* have the properties of analgesic, astringent, antispasmodic, antibacterial, carminative, contraceptive, demulcent, emollient, febrifuge, immunostimulant, laxative, stimulative, tonic, vermifuge and many more [11, 17, 23].
C. rotundus is used for stomach disorders, nausea, vomiting, intestinal parasites, food poisoning, indigestion, diarrhea, dysentery, irritation of the bowel and for curing amenorrhea, bronchitis, blood disorders, leprosy, menstrual irregularities, and inflammatory diseases [1, 2, 9]. Its whole plant extract is used as anti-nociceptive [10] and as a tonic for the liver and heart, a digestion stimulant, and aid against hypertension [4]. The rhizomes and tubers of *C. rotundus* have antioxidant, anti-inflammatory, antimutagenic, antiperiodic, anticonvulsant, anti-saturative, antipyretic, antibacterial, antiviral, antifungal, antidiabetic, antimalarial, antilipidemic, anti-tumoral, cardioprotective, and wound-healing properties [15, 18, 22, 25]. Alkaloids, tannins, essential oils (α-longipinane, β-selinene, cyperene, and caryophyllene oxide), and flavonoids such as anthocyanidins, catechins, flavans, flavones, flavanonols, and isoflavane are the active ingredients responsible for the biological and therapeutic properties of *C. rotundus* [8, 18].

Photo plate 1: *Cyperus rotundus* L plant.

Photo plate 2: *C. Rotundus* dried aerial parts

Photo plate 3: *C. Rotundus* dried underground parts

Materials and Methods

Collection of plant material: The aerial and underground parts of *C. rotundus* plants were collected from the field and near damp water of Degloor and Dharmabad, Nanded Dist, Maharashtra state.

Preparation of plants powder: The aerial and underground parts of *Cyperus rotundus* were washed separately with distilled water to remove any external debris. The washed parts were shade dried at room temperature. After complete drying, aerial and underground parts were ground to a fine powder using a blender, packed in an air tight container and stored in a refrigerator at 4ºC for further use.

Soxhlet extraction: 30 grams of both powdered samples was loaded to a blank thimble filter and covered with cotton. 300ml of ethanol was poured into the round bottom extraction flask and the thimble containing the sample was placed into the extraction chamber. The extraction was carried out. The flask containing solvent and extracted crude was taken after the extraction process [20].

Gas Chromatography-Mass Spectrometry (GCMS) analysis:

GC-MS analysis and characterization of ethanol extracts of aerial and underground parts of *C. rotundus* was done with EI- MS Spectrum scanned at 70 eV at SAIF, IIT Bombay. The relative percentage amount of each component was calculated by comparing its average peak area with the total area.

Instrument details

Agilent 7890 instrument was used for GC, detector used was Flame Ionization Detector (FID).

Joel Accu Time of Flight Analyzer (TOF) GCV instrument for MS was used, Specification: Mass range of 10-2000 amu and resolution is of 6000.

GC-MS analysis was performed by split less injection (80-1M-6-200-2M-8-275-5M-5-280-ETHANOL-HP5).

Results

The GC-MS spectrum of ethanolic extract of aerial parts of *Cyperus rotundus* shown in Fig 1. In GC-MS analysis total 19 bioactive compounds were identified. The identification of phytocompounds based on molecular formula and retention times (RT) being compared to the database of National Institute of Standards and Technology (NIST). The phytocompounds were identified as Cyclopropylmethyl carbinol; Vinyl butyrate; 4,5-Dihydro-β,β,4,4-tetramethyl-1H-pyrazole-1-propanol; Tricyclo[4.2.1.0(3,7)]nonane-3,8-diol, (anti-8)-; (+-)-3-Methyl-1-penten-3-ol; Hexyl isopropyl succinate; Succinic acid, pentyl tetrahydrofurfuryl ester; Oxetane, 2,4-dimethyl-, trans-; 2-Butanol, 2-methyl-, acetate; Formic acid, 2-methylpropyl ester; Ethanol, 2,2-dichloro-; Propane; Oxirane, (2-methylpropyl)-; Butanal, 4-[(tetrahydro-2H-pyran-2-yl)oxy]-; Phosphonofluoridic acid, (1-methylethyl)-, hexyl ester; Tridecane, 6-methyl-; (+-)-3-Methyl-1-penten-3-ol; Acetamide, 2-(2,3,6,7-tetrahydro-1,3,7-trimethyl-2,6-dioxo-1H-purin-8-ylthio)- and Sulfurous acid, nonyl 2-propyl ester as major compounds.

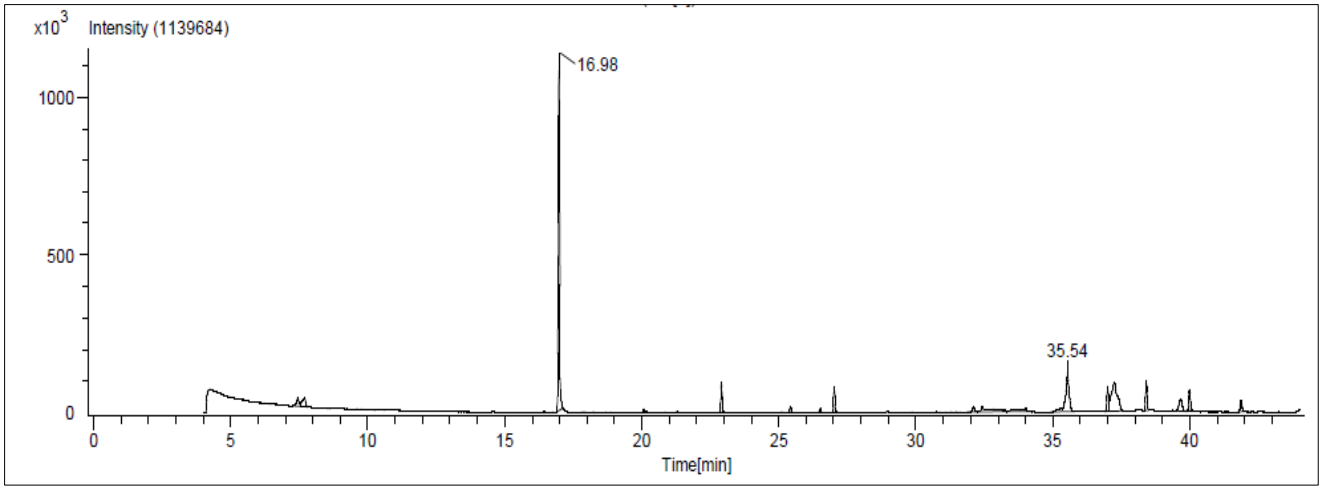

Fig 1: GC-MS spectrum of ethanolic extract of aerial parts of *Cyperus rotundus*.

Table 1: GC-MS analysis identified compounds of aerial parts of *Cyperus rotundus.*

Sr. No.	Compound name	Molecular formula	Molecular weight	RT (min)	Peak area (%)
1	Cyclopropylmethyl carbinol	$C_5H_{10}O$	86	7.45	1.84
2	Vinyl butyrate	$C_6H_{10}O_2$	114	7.70	2.67
3	4,5-Dihydro-β,β,4,4-tetramethyl-1H-pyrazole-1-propanol	$C_{10}H_{20}N_2O$	184	16.98	38.82
4	Tricyclo[4.2.1.0(3,7)]nonane-3,8-diol, (anti-8)-	$C_9H_{14}O_2$	154	20.06	0.22
5	(+-)-3-Methyl-1-penten-3-ol	$C_6H_{12}O$	100	20.18	0.10
6	Hexyl isopropyl succinate	$C_{13}H_{24}O_4$	244	22.91	3.32
7	Succinic acid, pentyl tetrahydrofurfuryl ester	$C_{14}H_{24}O_5$	272	25.42	0.65
8	Oxetane, 2,4-dimethyl-, trans-	$C_5H_{10}O$	86	26.51	0.34
9	2-Butanol, 2-methyl-, acetate	$C_7H_{14}O_2$	140	27.02	2.63
10	Formic acid, 2-methylpropyl ester	$C_5H_{10}O_2$	102	32.10	1.01
11	Ethanol, 2,2-dichloro-	$C_2H_4Cl_2O$	114	32.42	4.54
12	Propane	C_3H_8	44	34.01	2.42
13	Oxirane, (2-methylpropyl)-	$C_6H_{12}O$	100	35.54	11.8
14	Butanal, 4-[(tetrahydro-2H-pyran-2-yl)oxy]-	$C_9H_{16}O_3$	172	37.00	3.44
15	Phosphonofluoridic acid, (1-methylethyl)-, hexyl ester	$C_9H_{20}FO_2P$	210	37.24	14.5
16	Tridecane, 6-methyl-	$C_{14}H_{30}$	198	38.41	3.52
17	(+-)-3-Methyl-1-penten-3-ol	$C_6H_{12}O$	100	39.67	3.14
18	Acetamide, 2-(2,3,6,7-tetrahydro-1,3,7-trimethyl-2,6-dioxo-1H-purin-8-ylthio)-	$C_{10}H_{13}N_5O_3S$	283	39.99	3.01
19	Sulfurous acid, nonyl 2-propyl ester	$C_{12}H_{26}O_3S$	250	41.87	1.84

The GC-MS spectrum of ethanolic extract of underground parts of *Cyperus rotundus* is shown in Fig 2. The GC-MS analysis shows the presence of 21 different bioactive compounds namely Acetic acid, dichloro-, ethyl ester; Ethaneperoxoic acid, 1-cyano-1-[2-(2-phenyl-1,3-dioxolan-2-yl)ethyl]pentyl ester; Furan, 2,3,5-trimethyl-; Decaborane (14); Ethyl citrate; Pyridine, 3-(2-propenyloxy)-; 1,4-Methanocycloocta [d] pyridazine, 1,4,4a,5,6,9,10,10a-octahydro-11, 11-dimethyl-, (1α,4α,4aα,10aα)-; 2-Hexenoic acid, 5-methyl-, 4-methylphenyl ester; β-n-Propylether of 11-epi-dihydroartemisinin; 6H-[1] Benzothiopyrano [4,3-b] quinoline, 6,6,7-trimethyl-; Anthiaergostan-5,7,9,22-tetraen, 3-ureido-; 2,6-Heptanedione, 3-methyl-3-(1-methylethyl)-; 1,3(2H)-Benzodioxole, 2-[(diethylamino) methyl]-5-[(2,4-diamino-5-pyrimidinyl) methyl]-; Undecanoic acid, 2-methyl-, methyl ester; Ether, 3-butenyl pentyl; Propane, 1-iodo-2,2-dimethyl-; Butanethioic acid, 3-oxo-, S-butyl ester; Ether, 3-butenyl pentyl; 2-Ethyl-1-butanol, trifluoroacetate; Propanal, butylhydrazone and Ether, 3-butenyl propyl were identified.

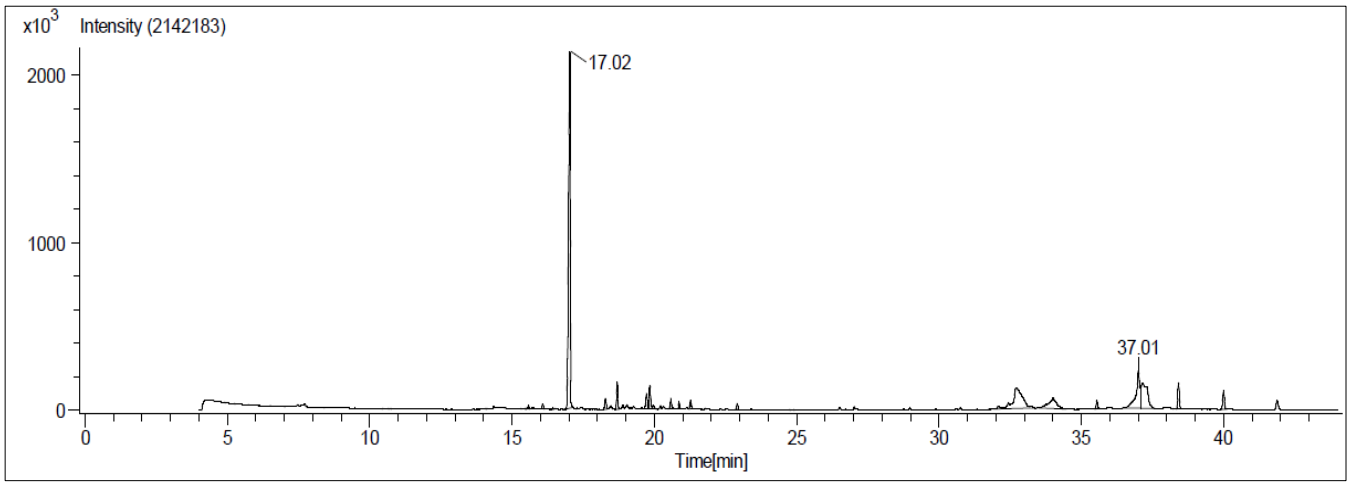

Fig 2: GC-MS spectrum of ethanolic extract of underground parts of *Cyperus rotundus.*

Table 2: GC-MS analysis identified compounds of underground parts *of Cyperus rotundus*.

Sr. No.	Compound name	Molecular formula	Molecular weight	RT (min)	Peak Area (%)
1	Acetic acid, dichloro-, ethyl ester	$C_4H_6Cl_2O_2$	156	15.57	0.29
2	Ethaneperoxoic acid, 1-cyano-1-[2-(2-phenyl-1,3-dioxolan-2-yl)ethyl]pentyl ester	$C_{19}H_{25}NO_5$	347	15.73	0.142
3	Furan, 2,3,5-trimethyl-	$C_7H_{10}O$	110	16.08	0.445
4	Decaborane(14)	$B_{10}H_{14}$	124	16.44	0.091
5	Ethyl citrate	$C_{12}H_{20}O_7$	276	17.03	42.00
6	Pyridine, 3-(2-propenyloxy)-	C_8H_9NO	135	18.28	1.02
7	1,4-Methanocycloocta[d]pyridazine, 1,4,4a,5,6,9,10,10a-octahydro-11,11-dimethyl-, (1α,4α,4aα,10aα)-	$C_{13}H_{20}N_2$	204	18.69	1.95
8	2-Hexenoic acid, 5-methyl-, 4-methylphenyl ester	$C_{14}H_{18}O_2$	218	19.72	1.40
9	β-n-Propylether of 11-epi-dihydroartemisinin	$C_{18}H_{30}O_5$	326	19.84	2.06
10	6H-[1]Benzothiopyrano[4,3-b]quinoline, 6,6,7-trimethyl-	$C_{19}H_{17}NS$	291	19.96	0.35
11	Anthiaergostan-5,7,9,22-tetraen, 3-ureido-	$C_{29}H_{44}N_2O$	436	20.58	1.00
12	2,6-Heptanedione, 3-methyl-3-(1-methylethyl)-	$C_{11}H_{20}O_2$	184	20.87	0.56
13	1,3(2H)-Benzodioxole, 2-[(diethylamino)methyl]-5-[(2,4-diamino-5-pyrimidinyl) methyl]-	$C_{17}H_{23}N_5O_2$	329	21.27	0.83
14	Undecanoic acid, 2-methyl-, methyl ester	$C_{13}H_{26}O_2$	214	22.91	0.48
15	Ether, 3-butenyl pentyl	$C_9H_{18}O$	142	32.71	14.2
16	Propane, 1-iodo-2,2-dimethyl-	$C_5H_{11}I$	198	34.02	5.95
17	Butanethioic acid, 3-oxo-, S-butyl ester	$C_8H_{14}O_2S$	174	35.54	0.77
18	Ether, 3-butenyl pentyl	$C_9H_{18}O$	142	37.01	9.65
19	2-Ethyl-1-butanol, trifluoroacetate	$C_8H_{13}F_3O_2$	198	37.16	11.74
20	Propanal, butylhydrazone	$C_7H_{16}N_2$	128	38.42	2.78
21	Ether, 3-butenyl propyl	$C_7H_{14}O$	114	40.00	2.20

Discussion

Since ancient times, plants have been utilised in traditional medical systems. The search for novel drugs has traditionally been guided by conventional knowledge of medicinal plants [3, 13]. There is emerging understanding of how to link the biological functions of phytochemical substances [7, 21, 24]. The therapeutic properties of plants are because of phytochemicals. Many pharmaceutical industries have been founded on the basis of this fundamental understanding. Using GC-MS, several phytochemical screening investigations have been conducted around the globe [27, 28].

The GC-MS analysis in the present study revealed 19 and 21 phytochemical compounds in aerial and underground parts of ethanolic extracts of *C. Rotundus*. The gas chromatogram showed that the relative concentration of various compounds getting eluted at different retention times. Sulfurous acid, nonyl 2-propyl ester (41.87 RT) is the highest retention time chemical compound and Cyclopropylmethyl carbinol (7.45) is the lowest retention time chemical compound in the aerial parts. Whereas in the underground parts Ether, 3-butenyl propyl (40.00 RT) is the highest retention time chemical compound and Acetic acid, dichloro-, ethyl ester (15.57) is the lowest retention time chemical compound. The heights of the peak indicate that the relative concentrations of the components present in the plant extract.

Conclusion

The ethanolic extract of *Cyperus rotundus* aerial and underground parts revealed the presence of 19 and 21 different phytochemical compounds based on their retention time, molecular weight, peak area and MS fragment ions generated. Each phytochemical compound can be extracted individually and can be used in clinical trials to evaluate effectiveness, and to develop a novel drug from a crude drug.

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Conflict of Interest

Authors declare that there is no conflict of interest.

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