



GAS CHROMATOGRAPHY-MASS SPECTROMETRIC ANALYSES OF ACETONE EXTRACT OF MARWAR DHAMAN GRASS FOR BIO-ACTIVE COMPOUNDS

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Abstract

The investigation was carried out to determine the possible bioactive components of *Cenchrus setigerus* using Gas chromatography-Mass spectrometry (GC-MS). All the samples were dried firstly at 60°C for 2 days in an oven after that leave it on room temperature. They were then macerated to powder form with a mixer grinder. The powder was stored in air sealed polythene bags at room temperature before extraction. The chemical compositions of the acetone extract of *C. setigerus* were investigated using Perkin-Elmer Gas chromatography-Mass spectrometry, while the mass spectra of the compounds found in the extract was matched with the National Institute of Standards and Technology (NIST) and WILEY 8 library. Total 67 components of the above said plant were identified. This is the first report of identification of components from the whole plant of *C. setigerus* by GC-MS.

Key words : *Cenchrus setigerus*, GC-MS analysis, bioactive components and acetone extract.

Introduction

Taking into consideration of the medicinal importance of this plant, the acetone extract of the whole plant of *Cenchrus setigerus* (Marwar Dhaman) was analyzed for the first time using Gas chromatography-Mass spectrometry (GC-MS). This work will help to identify the compounds of therapeutic value. GC-MS is one of the best techniques to identify the bioactive constituents of long chain, branched chain hydrocarbons, alcohols, acids, ester, steroids, phenolic compounds etc. (Amakrishnan, 2011).

Medicinal plants are used in traditional treatments to cure variety of diseases. In the last few decades there has been an exponential growth in the field of herbal medicine. Natural products have been a source of drugs for centuries. In the present study, acetone extract of *C. setigerus* was analyzed by GC-MS technique to study the major and minor phyto-constituents of the vegetative parts of the whole plant.

Cenchrus setigerus is extremely variable species Tufted, non-rhizomatous (or shortly rhizomatous), erect

or ascending perennial to 60 cm (rarely to >1 m) tall, perennial, with types ranging in habit from ascendant to erect, and branching culms from about 0.3-2.0m at maturity. Common names of this grass are Birdwood grass, cow sandbur (English), moda dhaman grass, motha dhaman, kata-dhaman, kala-dhaman (India). Leaf blades are linear, 15-30 cm long, and 4-6 mm wide. Panicle a green, maturing to light straw-coloured (mostly African origin) or dark purple (mostly Indian origin), false spike, 1.5-9 cm long and ca. 1 cm wide. Seed units or fascicles are inserted along a zig-zag axis, each bur-like fascicle comprising a single spikelet or cluster of 2 or 3 spikeletes, 3-4.5 mm long. 180,000-350,000 seed units/kg. This grass distributed throughout Africa, Arabia and India. It is adventive in the southern United States, Australia and South America where, it has been introduced as an experimental forage grass. Special characters of *Cenchrus setigerus* (Marwar Dhaman grass) (CAZRI-76) is selected from exotic material and well adopted in the arid and semi-arid regions of India. 'Marwar Dhaman' is excellent for grazing purpose due to its thin stem and leafy foliage. It is a drought hardy perennial grass, which forms clumps at the base. It is an early maturing variety,

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flowers between 45 to 55 days, high in tillering, with good regeneration abilities and capable of giving 2 to 3 cuts per year under rainfed conditions. It provides an average yield of 40 quintal/hectare green fodder and 5 quintal/hectare dry matter in desertic region, whereas in semi-desertic areas, the yield becomes doubled. It contains 9.5% crude protein and has 65% digestibility at half bloom stage. Its pasture remains productive for 4 to 5 years. It is moderately resistant to major insect pests (Anonymous, 1988).

This grass is gaining attention in various field of research, as they are best suited to the present environmental conditions (Singariya *et al.*, 2012a). This is more competitive under the conditions of high temperature (Singariya *et al.*, 2012u), solar radiation and low moisture (Agrawal, 2007) and is more efficient at gathering CO₂ and utilizing nitrogen from the atmosphere and recycled N in the soil (Bessman, 1956 and Singariya, 2009). This grass has excellent soil binding capacity, which helps to conserve soil in desert areas (Sinha *et al.*, 1996). This grass, fed green, turned into silage, or made into hay is said to increase flow of milk in cattle and impart a sleek and glossy appearance (Singariya *et al.*, 2011a). Seeds of this grass are used as famine food by the tribal during severe conditions (Katewa and Sharma, 2004). However, *C. setigerus* is most suitable and highly nutritive grasses for desert environmental conditions (Singariya *et al.*, 2012b).

Materials and Methods

Plant material

Cenchrus setigerus (CAZRI -76) were collected in the month of August 2009 from the Central arid zone research institute CAZRI, Jodhpur (Rajasthan), India. Plants samples were identified and deposited in the herbarium, Department of Botany, University of Rajasthan, Jaipur (Rajasthan), India. The collected plant materials were transferred immediately to the laboratory cleaned with water and selected plant parts were separately shade dried until weight has been constant (Singariya *et al.*, 2012l).

Preparation of plant extracts

Preparation of plant extracts : The collected plant materials were shade dried, powered with the help of grinder (Singariya *et al.*, 2012m) and passed through 40mm meshes and stored in clean container for further use (Singariya *et al.*, 2012o). The dried powder material was extracted with acetone by using the Soxhlet apparatus (Subramanian and Nagarajan, 1969) for 18 hours at a temperature not exceeding the boiling point of the respective solvent (Singariya *et al.*, 2013 and Singariya

et al., 2012k). The obtained extracts were filtered by using Whatman No. 1 filter paper and then concentrated at 40°C by using an evaporator (Singariya *et al.*, 2012p) and stored the residual extracts in refrigerator at 4°C in small and sterile amber colour glass bottles (Singariya *et al.*, 2012n) for subsequent use in the further antimicrobial, anti-fungal and phyto-chemical analysis. The extract contains both polar and non-polar phyto-components.

Gas chromatography-Mass spectrometry analysis

Gas chromatography-Mass spectrometry (GC-MS) analysis of these extracts was carried out by following the method of Hema *et al.* (2010). The GC-MS analysis of the extracts was performed using a GC-MS (Model; QP 2010 series, Shimadzu, Tokyo, Japan) equipped with a VF-5ms fused silica capillary column of 60m length, 0.25mm dia. and 0.25mm film thickness (Singariya *et al.*, 2014). Injection Mode: Split, Flow Control Mode: Linear Velocity, Pressure: 173.3 kPa, Linear Velocity: 28.9cm/sec, Purge Flow: 3.0 mL/min, Split Ratio: 10.0. For GC-MS detection [GC-2010], an electron ionization system with ionization energy of 70eV was used. Helium gas (99.99%) was used as a carrier gas at a constant flow rate- total flow: 16.3 mL/min. and column flow: 1.21 mL/min. injector and mass transfer line temperature were set at 200 and 240°C respectively. The oven temperature was programmed (Column Oven Temp.: 100.0°C and Injection Temp.: 270.00°C) from 70 to 220°C at 10°C/min, held isothermal for 1 min and finally raised to 300°C AT 10°C/min. 2ml of respective diluted samples was manually injected in the split less mode, with split ratio of 1:40 and with mass scan of 50-600 amu. Total running time of GC-MS is 48 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 Turbomass. The relative percentage of the each extract constituents was expressed as percentage with peak area normalization.

Identification of components

Interpretation on mass spectrum of GC-MS was done using the database of National Institute of standard and Technology NIST-08 LIB. (Mc-Lafferly, 1989) and WILEY-8 LIB. (Stein, 1990) library sources were used for matching the identified components from the plant material having more than 62,000 patterns. The mass spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight and structure of the components of the test materials were ascertained (Singariya *et al.*, 2012w).

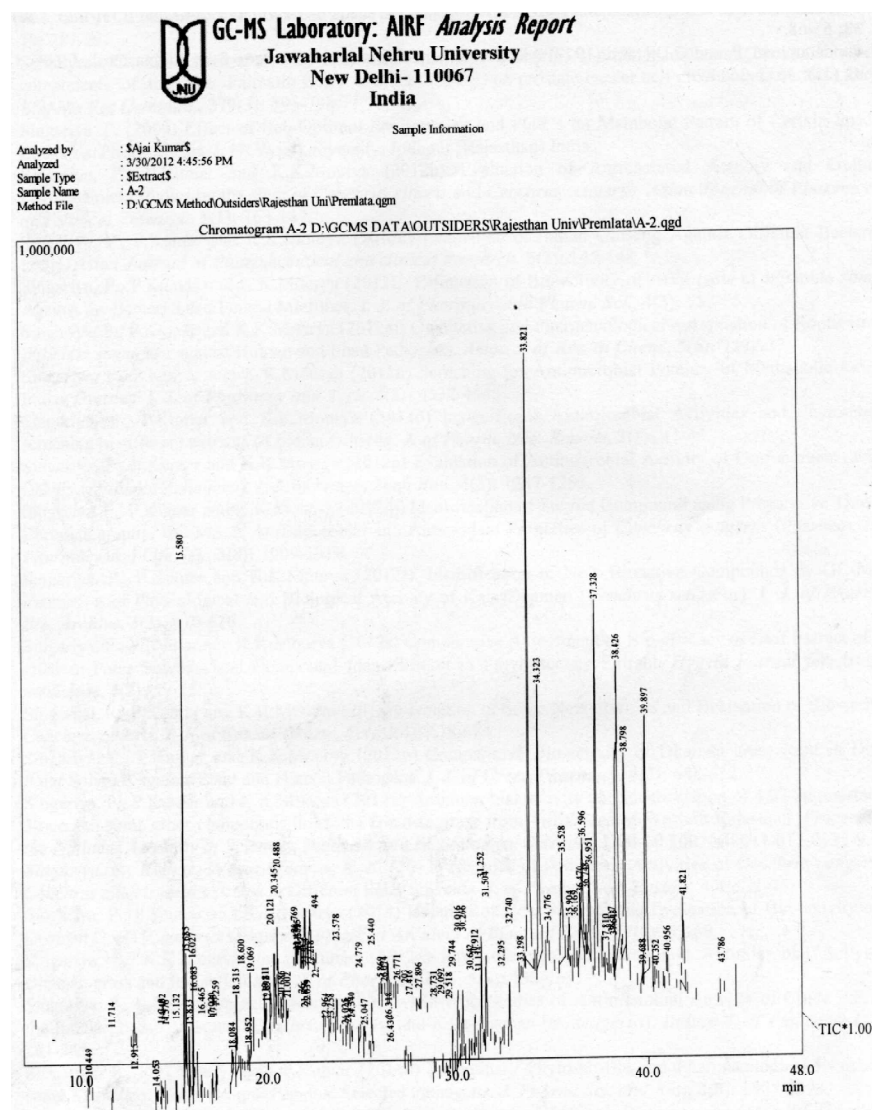


Fig. 1 : Chromatogram of Acetone extract of *Cenchrus setigerus* by GC-MS.

Results and Discussion

The active principles with their retention time (RT), molecular formula (MF), molecular weight (MW) and concentration (%) in the acetone extracts of the whole plant of *C. setigerus* are presented in tables 1 and 2 (Singariya *et al.*, 2012q). The GC-MS analysis of the extracts showed the presence of phyto-components, the phyto-components of the above said plant extract are presented in table 1 and the GC-MS chromatogram with peak area of each extract is also given fig. 1. Fatty acid ethyl esters perturb the cell cycle and induce apoptosis in the cancer cells (Aydin *et al.*, 2005). They are also used as markers of excessive alcohol consumption (Hartwig *et al.*, 2003). Hexadecenal is a prevalent component found in many medicinal plants (Nazlina *et al.*, 2011). Alpha-tocopherol has potent anti-oxidant properties. A reduction of 34% is observed in the incidence of prostate

cancer in smokers given daily supplements of 50 mg of alpha-tocopherol. Other analysis also suggests the association of alpha-tocopherol (vitamin E) supplement use with a 15% lower risk of prostate cancer (Heinonen *et al.*, 1998).

Totally 67 bio-active constituents were identified in the present study from the acetone extracts of the whole plant of *C. setigerus*, which including both major and minor constituents. In previous investigation, Cholest-4-en-3-one (2.36%) was reported in acetone extract of *C. setigerus* (Singariya *et al.*, 2012r). Ethyl iso-allocholate has antimicrobial, Diuretic and anti-inflammatory activity (Singariya *et al.*, 2012t). 4,22-stigmastadiene-3-one (2.41%) were found in the ethyl acetate extract of *C. ciliaris* (Singariya *et al.*, 2012s). The major ten constituents were (fig. 2 : The best hits for the prevailing compounds in the chromatogram) (14.39%) 9,19-Cycloergost-24(28)-

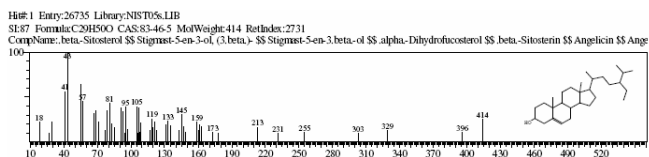
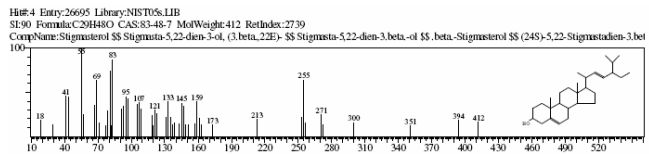
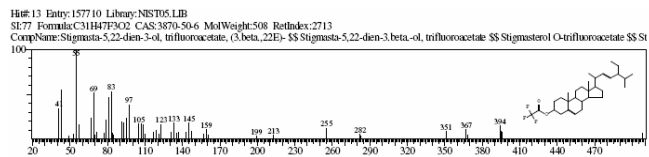
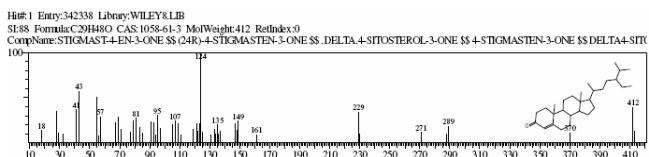
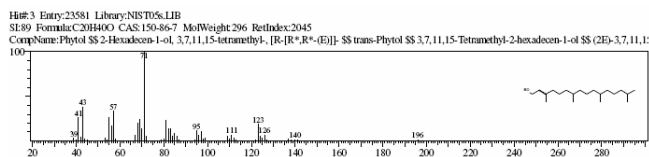
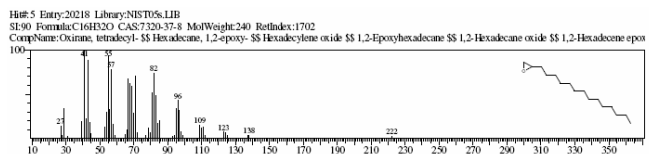
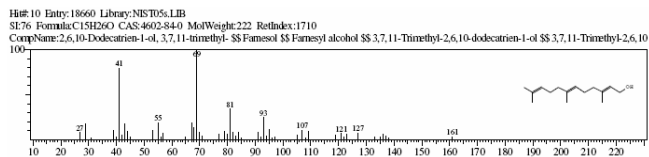
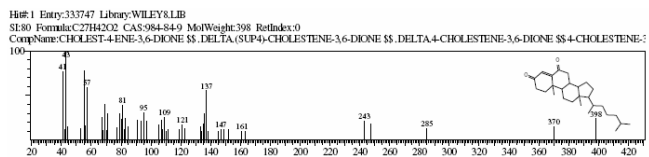
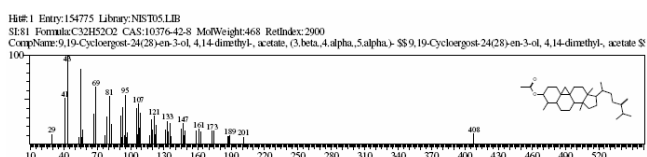
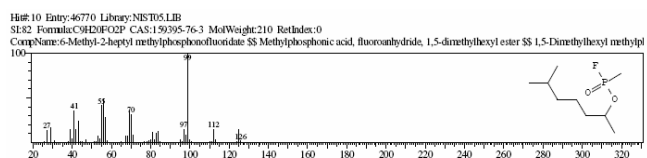
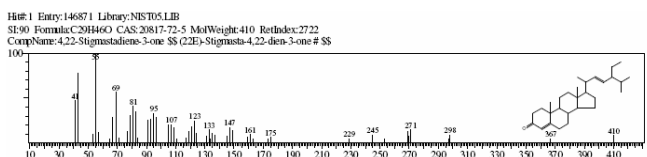
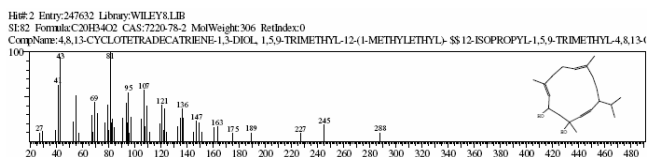
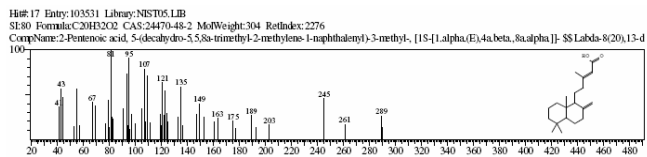
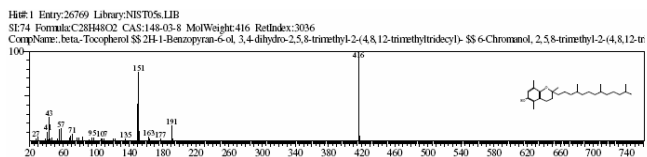
**Beta-Sitosterol****Stigmastol****Stigmast-5,22-dien-3-ol, trifluoroacetate, (3.beta.,22E)-****Stigmast-4-en-3-one****Phytol****Oxirane, tetradecyl-****2,6,10-Dodecatrien-1-ol,3,7,11-trimethyl-****Cholest-4-ene-3,6-dione****9,19-Cycloergost-24(28)-en-3-ol,4,14-dimethyl-, acetate,(3.beta.,4.alpha.,5.alpha.)-****6-Methyl-2-heptyl methylphosphonofluoridate****4,22-Stigmastadiene-3-one****4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-Trimethyl-12-(1-Methylmethyl)-****2-Pentenoic acid, 5-(decahydro-5,5,8a-trimethyl-2-methylene-1-naphthalenyl)-3-methyl-, [1S-[1.alpha.(E),4.alpha.(E),8.alpha.(E)]]-****Beta-Tocopherol****Fig. 2 : The best hit for the prevailing compounds in the chromatogram.**

Table1: Compounds identified in the acetone extract of *Cenchrus setigerus*.

P	R. T.	Area	Area (%)	Compound name	M. F.	M. W.
1	10.449	40318	0.16	Butane, 2,2-dimethyl-	C ₆ H ₁₄	86
2	11.714	30602	0.12	Hexadecane	C ₁₆ H ₃₄	226
3	12.915	64208	0.26	Pentane, 2,2-dimethyl-	C ₇ H ₁₆	100
4	14.053	29640	0.12	Nonane, 3,7-dimethyl-	C ₁₁ H ₂₄	156
5	14.348	18519	0.07	1,7-Heptanedio	C ₇ H ₁₆ O ₂	132
6	14.402	16886	0.07	1-Octanol, 3,7-dimethyl-	C ₁₀ H ₂₂ O	158
7	14.576	27451	0.11	3-[(Z)-1-Butenyl]-4-vinylcyclopentene	C ₁₁ H ₁₆	148
8	15.132	38591	0.16	Dodecane, 1,1-difluoro-	C ₁₂ H ₄₂ F ₂	206
9	15.580	1009148	4.07	Oxirane, tetradecyl-	C ₁₆ H ₃₂ O	240
10	15.685	213271	0.86	2-Pentadecanone, 6,10,14-trimethyl-	C ₁₈ H ₃₆ O	268
11	15.833	146458	0.59	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	C ₂₀ H ₄₀ O	296
12	16.027	264109	1.07	1-Octadecyne	C ₁₈ H ₃₄	250
13	16.083	39911	0.16	Phthalic acid, butyl undecyl ester	C ₂₃ H ₃₆ O ₄	376
14	16.465	32161	0.13	Methyl cyclohexanepropionate	C ₁₀ H ₁₈ O ₂	170
15	17.259	45383	0.18	Hexadecanoic acid, 2-oxo-, methyl ester	C ₁₇ H ₃₂ O ₃	284
16	18.084	130132	0.53	4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester	C ₁₃ H ₁₅ NO ₃	233
17	18.315	314880	1.27	Phytol	C ₂₀ H ₄₀ O	296
18	19.069	169291	0.68	1-Pentadecanol acetate	C ₁₇ H ₃₄ O ₂	270
19	19.811	68128	0.27	1,6-Octadien-3-ol, 3,7-dimethyl-	C ₁₀ H ₁₈ O	154
20	19.892	37345	0.15	9,12,15-Octadecatrienoic acid, 2-(acetyloxy)-1-[(acetyloxy)methyl] ethyl ester, (Z,Z,Z)-	C ₂₅ H ₄₀ O ₆	436
21	20.121	108133	0.44	Myristaldehyde	C ₁₄ H ₂₈ O	212
22	20.345	142411	0.57	Cyclobutanecarboxylic acid, undec-2-enyl ester	C ₁₆ H ₂₈ O ₂	252
23	20.488	289774	1.17	6-Methyl-2-heptyl methylphosphonofluoridate	C ₉ H ₂₀ FO ₂ P	210
24	20.769	44961	0.18	1-Heptadecanol, acetate	C ₁₉ H ₃₈ O ₂	298
25	21.934	115396	0.47	Palmitaldehyde	C ₁₆ H ₃₂ O	240
26	22.218	225804	0.91	Di-n-octyl phthalate	C ₂₄ H ₃₈ O ₄	390
27	23.578	93567	0.38	Sulfurous acid, 2-ethylhexyl isohexyl ester	C ₁₄ H ₃₀ O ₃ S	278
28	24.779	41196	0.17	13-Octadecenal, (Z)-	C ₁₈ H ₃₄ O	266
29	25.440	153583	0.62	Squalene	C ₃₉ H ₅₀	410
30	25.869	39936	0.16	Farnesol	C ₁₅ H ₂₆ O	222
31	26.074	51903	0.21	2-Bromopropionic acid, 6-ethyl-3-octyl ester	C ₁₃ H ₂₅ BrO ₂	292
32	26.318	108133	0.44	Tetratetracontane	C ₄₄ H ₉₀	618
33	26.771	52528	0.21	3-Keto-isosteviol	C ₂₀ H ₂₈ O ₄	332
34	27.896	49973	0.20	8-Hexadecenal, 14-methyl-, (Z)-	C ₁₇ H ₃₂ O	252
35	28.731	74890	0.30	Cholesta-8,24-dien-3-ol, 4-methyl-, (3β, 4α)-	C ₂₈ H ₄₆ O	396
36	29.092	86028	0.35	Ethyl iso-allocholate	C ₂₆ H ₄₄ O ₅	436
37	29.744	156093	0.63	β-Carotene	C ₄₀ H ₅₆	536
38	30.016	513025	2.07	β-Tocopherol	C ₂₈ H ₄₈ O ₂	416
39	30.266	368054	1.48	Heptacosane	C ₂₇ H ₅₆	380
40	30.642	101422	0.41	2,6,10,14-Hexadecatetraenoic acid, 3,7,11,15-tetramethyl-, methyl ester, (E,E,E)-	C ₂₁ H ₃₄ O ₂	318

Table 1 continued.....

Table 1 continued.....

41	30.912	107172	0.43	Cholesta-4,6-dien-3-ol, (3 β)-	C ₂₇ H ₄₄ O	384
42	31.114	73849	0.30	Oxalic acid, allyl pentadecyl ester	C ₂₀ H ₃₆ O ₄	340
43	31.252	378163	1.53	3 α , 7 β -Dihydroxy-5 β , 6 α -epoxycholestane	C ₂₇ H ₄₆ O ₃	418
44	31.504	334150	1.35	Vitamin E (α -Tocopherol)	C ₂₉ H ₅₀ O ₂	430
45	32.295	87594	0.35	(E,E,E)-3,7,11,15-Tetramethylhexadeca-1,3,6,10,14-pentaene	C ₂₀ H ₃₂	272
46	32.740	205462	0.83	10-12-Pentacosadiynoic acid	C ₂₅ H ₄₂ O ₂	374
47	33.298	69642	0.28	Kauran-18-al, 17-(acetyloxy)-, (4 β)-	C ₂₂ H ₃₄ O ₃	346
48	33.821	3567706	14.39	9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethyl-, acetate, (3 β , 4 α , 5 α)-	C ₃₂ H ₅₂ O ₂	468
49	34.323	1741978	7.03	Stigmasterol	C ₂₉ H ₄₈ O	412
50	34.776	216652	0.87	Cholestan-3-one, (5 α)-	C ₂₇ H ₄₆ O	386
51	35.528	932218	3.76	β -Sitosterol	C ₂₉ H ₅₀ O	414
52	35.904	174483	0.70	Acetic acid, 3-hydroxy-7-isopropenyl-1, 4a-dimethyl-2,3,4,4a,5,6,7,8-octahydronaphthalen-2-yl ester	C ₁₇ H ₂₆ O ₃	278
53	36.169	169755	0.68	Longifolenaldehyde	C ₁₅ H ₂₄ O	220
54	36.470	159975	0.65	Ergosta-7,22-dien-3-ol, (3 β , 22E)-	C ₂₈ H ₄₆ O	396
55	36.596	584906	2.36	Cholest-4-en-3-one	C ₂₇ H ₄₄ O	384
56	36.748	130812	0.53	Cholestane-2,3-diol, 3-acetate	C ₂₉ H ₅₀ O ₃	446
57	36.951	504754	2.04	Lupeol (Fagarsterol)	C ₃₀ H ₅₀ O	426
58	37.328	2111192	8.52	4,22-Stigmastadiene-3-one	C ₂₉ H ₄₆ O	410
59	37.613	287194	1.16	2(1H)Naphthalenone, 3,5,6,7,8,8a-hexahydro-4,8a-dimethyl-6-(1-methylethenyl)-	C ₁₅ H ₂₂ O	218
60	37.810	156012	0.63	Stigmasta-3,5-dien-7-one	C ₂₉ H ₄₆ O	410
61	38.117	77638	0.31	Ergosta-4,6,22-trien-3-one	C ₂₈ H ₄₂ O	394
62	38.426	2738166	11.05	2-Pentenoic acid, 5-(decahydro-5,5,8a-trimethyl -2-methylene-1-naphthalenyl)-3-methyl-, [1S-[1 α (E), 4 α β , 8 α α]]-	C ₃₀ H ₃₂ O ₂	304
63	38.798	1290631	5.21	Stigmast-4-en-3-one	C ₂₉ H ₄₈ O	412
64	39.897	1893135	7.64	4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12-(1-methylethyl)-	C ₂₀ H ₃₄ O ₂	306
65	40.956	236905	0.96	Formic acid, 3,7,11-trimethyl-1,6,10-dodecatrien-3-yl ester	C ₁₆ H ₂₆ O ₂	250
66	41.821	761365	3.07	Stigmasta-5,22-dien-3-ol, trifluoroacetate, (3 β , 22E)-	C ₃₁ H ₄₇ F ₃ O ₂	508
67	43.786	241151	0.97	Cholest-4-ene-3,6-dione	C ₂₇ H ₄₂ O ₂	398
		24785902	100.00			

en-3-ol, 4,14-dimethyl-, acetate, (3 β , 4 α , 5 α)-; (11.05%) 2-Pentenoic acid, 5-(decahydro-5,5,8a-trimethyl -2-methylene-1-naphthalenyl)-3-methyl-, [1S-[1 α (E), 4 α β , 8 α α]]-, (8.52%) 4,22-Stigmastadiene-3-one; (7.64%) 4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12-(1-methylethyl)-; (7.03%) Stigmasterol; (5.21%) Stigmast-4-en-3-one; (4.07%) Oxirane, tetradecyl-; (3.76%) β -Sitosterol; (3.07%) Stigmasta-5,22-dien-3-ol, trifluoroacetate, (3 β , 22E)-; (2.36%) Cholest-4-en-3-one; (2.07%) β -tocopherol; (2.04%) Lupeol (Fagarsterol) and along with major constituents, minor constituents were (0.12%) Hexadecane; (0.12%) Nonane, 3,7-dimethyl-; (0.11%) 3-[(Z)-1-Butenyl]-4 vinylcyclopentene; (0.07%) 1,7-Heptanedio; (0.07%) 1-Octanol, 3,7-dimethyl-

Among the identified phytochemicals (tables 1 and 2) squalene has the property of antioxidant (Kala *et al.*, 2011). Recently squalene possesses chemopreventive activity against colon carcinogenesis (Rao *et al.*, 1998). Phytol is detected in *Polygala javana* whole plant, which was also found to be effective at different stages of the arthritis (Alagammal *et al.*, 2012). It was found to give food as well as preventive and therapeutic results against arthritis. The results show that, reactive oxygen species-promoting substances such as phytol constitute a promising novel class of pharmaceuticals for the treatment of rheumatic arthritis and possibly other chronic inflammatory diseases (Ogunlesi *et al.*, 2009).

Table 2 : Nature and activity of bio-active compounds identified in the Acetone extract of *Cenchrus setigerus*.

P	R. T.	Compound name	Nature of Compound	Activity
1	6.203	Pantolactone		
2	7.966	Hexylene Glycol	Alcoholic compound	Antimicrobial Preservative
3	14.047	Oxalic acid, isobutyl pentyl ester		
4	16.081	Phthalic acid, butyl undecyl ester		
5	17.253	Hexadecanoic acid, 2-oxo-, methyl ester		
6	17.383	Decanoic acid, propyl ester		
7	18.318	Phytol	Diterpene	Antimicrobial, Anti-inflammatory, Anticancer, Diuretic 5
8	20.119	Myristaldehyde		
9	20.350	Cyclobutanecarboxylic acid, undec-2-enyl ester		
10	21.532	Sulfurous acid, 2-ethylhexyl isohexyl ester		
11	21.937	Palmitaldehyde		
12	22.222	Di-n-octyl phthalate	Plasticizer compound	Antimicrobial Antifouling
13	25.440	Squalene	Triterpene	Antibacterial, Antioxidant, Anti-tumor, Cancer preventive, Immunostimulant, Chemo preventive, Lipoxxygenase-inhibitor, Pesticide Diuretic
14	26.769	3-Keto-isosteviol	Alcoholic compound	Antimicrobial
15	28.100	Oxalic acid, 6-ethyloct-3-yl heptyl ester		
16	29.093	Ethyl iso-allocholate	Steroid	Antimicrobial, Diuretic, Anti-inflammatory, Anti-asthma
17	29.752	β -Carotene		
18	30.015	β -Tocopherol		
19	30.908	Cholesta-4,6-dien-3-ol, (3 β)-	Steroid	Antimicrobial, Diuretic, Anti-inflammatory, Anti-asthma
20	31.259	Stigmasta-5,22-dien-3-ol, acetate, (3 β)-		
21	31.506	Vitamin E (α -Tocopherol)		
22	33.573	Pentafluoropropionic acid, heptadecyl ester		
23	33.702	Campesterol	Steroid	Anti-tumor, Cancer preventive, inhibit intestinal cholesterol absorption, Anti-inflammatory
24	34.340	Stigmasterol	Steroid	Anti-tumor, Cancer preventive, inhibit intestinal cholesterol absorption. Anti-inflammatory
25	35.545	β -Sitosterol	Steroid	Anti-tumor, Cancer preventive, inhibit intestinal cholesterol absorption. Anti-inflammatory
26	36.184	Longifolenaldehyde		
27	36.473	Ergosta-7,22-dien-3-ol, (3.beta.,22E)-		
28	36.582	Cholest-4-en-3-one		
29	36.743	Cholestane-2,3-diol, 3-acetate		
30	36.970	Lupeol (Fagarsterol)	Steroid	Anti-tumor, Cancer preventive, inhibit intestinal cholesterol absorption. Anti-inflammatory

Progesterone also inhibits the conversion of testosterone to dihydrotestosterone and stimulates the activity of p53, thereby finds application in prostate cancer therapy (Mercola, 1998 and South, 2012). Research has indicated that stigmasterol may be useful in prevention of certain cancers, including ovarian, prostate, breast and colon cancers. Studies with laboratory animals fed with stigmasterol suggest a decrease of 23% in the cholesterol absorption over a 6-week period. It inhibits several pro-inflammatory and matrix degradation mediators typically involved in osteoarthritis-induced cartilage degradation (Gabay *et al.*, 2010). It also exhibits potent antioxidant, hypoglycemic and thyroid inhibiting properties (Panda *et al.*, 2009). Beta-sitosterol and stigmasterol inhibit prostate cancer growth by increasing p53 protein expression and also inhibit carcinoma development by decreasing p21 and p27 protein expression (Scholtyssek *et al.*, 2009). Gamma-Sitosterol is a C-24 isomer of beta-Sitosterol (Grae me, 1963). According to Duke's ethanobotanical and phytochemistry database (Jim Duke, 1998), beta-Sitosterol and amyryn derivative, 12-Oleanen-3-yl acetate, (3 α) possess antioxidant, anti-inflammatory and antitumor activities.

The GC-MS chromatogram with peak area has shown in fig. 1. The aim of the present study is to provide more information about the essential phyto-constituents of *C. setigerus*. The results from the present investigation were very encouraging and indicates that this plant should be studied more extensively to explore its potential to use as plant medicinal nutritive.

Numerous studies have also reported the beneficial effects of the dietary intake of phytosterols. Ergost-5-en-3 beta-ol, also called "campesterol" competes with cholesterol and thus reduces the absorption of cholesterol in the human intestine (Heggen *et al.*, 2010; Choudhary and Tran, 2011). Phytosterols indirectly (*in-vivo* as a dietary supplement) and directly (in tissue culture media) inhibit the growth and metastasis of prostate cancer PC-3 cells (Awad *et al.*, 2001). In most of the parameters, beta-Sitosterol seems to be more effective than campesterol in offering this protection. Stigmasterol is used as the precursor of vitamin D3 (Kametani and Furuyama, 1987) and in the manufacture of synthetic progesterone, a valuable human hormone that plays an important physiological role in the regulatory and tissue building mechanisms related to estrogen effects, as well as acting as an intermediate in the biosynthesis of androgens, estrogens and corticoids (Sundararaman and Djerassi, 1977).

Conclusion

In the present study, 67 components from the acetone extracts of the whole plant of *C. setigerus* were identified by GC-MS analysis. The presence of various bioactive compounds justifies the use of this plant for various ailments by traditional practitioners. However, isolation of individual photochemical constituents and subjecting it to biological activity will definitely give fruitful results. It could be concluded that, *C. setigerus* contains various bioactive compounds. So, it is recommended as a plant of phyto-pharmaceutical importance. However, further studies are needed to undertake its bioactivity and toxicity profile.

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