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GC-MS ANALYSIS OF BIOACTIVE COMPOUNDS FROM THE METHANOLIC EXTRACT OF WHOLE PLANT OF *CYNODON DACTYLON* (L.) PERS.

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Cynodon dactylon (L), GC-MS analysis, Levoglucosenone, Dodecanoic acid, Thiophene, Hexadecanoic acid

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ABSTRACT: Plants have been an important source of medicine with qualities for thousands of years. The therapeutic effects in the plants are due to the chemical compounds there in. *Cynodon dactylon* (L) Pers commonly known as Bermuda grass belongs to the family Poaceae. In ethnomedicinal practices, the plant *Cynodon dactylon* used in the treatment of various diseases and has pharmacological actions. Its health and nutritional benefits are well documented but very little about its bioactive compounds. The present study was carried to identify the phyto-constituents present in the methanolic extract of *Cynodon dactylon* (L.) Pers by GC-MS analysis to ascertain its usage by the local community as a plant possessing medicinal properties. In total 68compounds were identified. The major constituents were furfural (2.33%), 3-aminopyrazine 1-oxide levoglucosenone (1.39%) tetetrapentacontane (2.47%), 9-octadecanoic acid (2.27%), n- hexadecanoic acid (7.5%), Neophytadiene (1.30%) and benzene propanoic acid (1.91%). The above findings supported the traditional use of *Cynodon dactylon* in various disorders.

INTRODUCTION: Medicinal plants are at great interest to drug industries, as herbal medicines and their derivative products are often prepared from crude plant extracts, which comprise a complex mixture of different phytochemical constituents. Development of herbal remedies is more popular now a day due to less side effects and easy availabilities of medicinal plants. Scientific and reliable reports indicated that about 25% of prescribed medicines worldwide are taken from herbs [1, 2].

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A substantial body of the literature has reported the multitude pharmacological applications of plant extracts and the compounds isolated from plant extracts [3]. Harnessing the biological potential of medicinal plants represents a sterling opportunity for the development of novel therapeutic candidates [4, 5]. The bioactive plant extracts are a promising source of many drugs. For example, berberine (Berberis) and Quinine (Cinchona) are the antibiotics obtained from plants, which are highly effective against bacteria (Escherichia coli and Staphylococcus aureus) [6].

Cynodon dactylon is commonly known as bermuda grass, belongs to family Poaceae. The plant is native to East Africa, Asia, Australia and Southern Europe. *Cynodon dactylon* has various medicinal properties. The plant is traditionally used as an agent to control diabetes. The extract of plant has

been reported to have potential antidiabetic, antioxidant & hypolipidemic efficacy. The plant possesses antiviral and antimicrobial activity too. The plant is astringent, sweet, cooling, haemostatic, depurative, vulnerary constipating, diuretic and tonic [6, 7]. Studies on lab animals have shown that methanolic extract of *C. dactylon* decreases the level of lipid peroxides. It was also revealed that the methanolic extract of *C. dactylon* had an antioxidant effect on COLO 320 DM cells, a colon cancer cell line, and the levels of antioxidant enzymes. Few studies have been conducted on medicinal and antimicrobial properties of *C. dactylon* (L.) Pers., and further studies must be carried out to confirm its antioxidant and antimicrobial properties. Chemical constituents of the methanolic extract of the whole plant of *C. dactylon* (L.) Pers [7]. From Sangrur district, Punjab were identified in this study for the first time and its potential activities were investigated. We evaluated the phytochemicals constituents of methanolic extract of *Cynodon dactylon* by gas chromatography and Mass spectrometry (GC-MS) to provide the scientific information to develop potential phytomedicine.

MATERIAL AND METHODS:

Materials: The whole plant of *Cynodon dactylon* was collected from the surrounding areas of Akal college of Pharmacy and Technical Education, Mastuana Sahib Sangrur, in the month of November2021. The plant was authenticated from CSIR-NIScPR, New Delhi having authentication no. NISc PR/RHMD/Consult 2021/3890-91.

Methods:

Preparation of Extract of *C. dactylon* Whole Plant: Whole plant of *Cynodon dactylon* was shade dried and coarsely powdered. The powdered plant material (500 g) was treated with various different solvents and extraction was performed by successive solvent extraction method. The extracts obtained were filtered & concentrated by using rota evaporator. From these extracts, methanolic extract was analyzed by GCMS.

GC-MS Analysis of Prepared Extract: GC-MS technique was used in this study to identify the phytocomponents present in the extract. The methanol extract of *Cynodon dactylon* was subjected to GC-MS detection. The detection was carried out with Gas chromatograph coupled with Mass spectrophotometer (GC-MS, Shimadzu QP 2010 Mass spectrophotometer). Helium was employed as the carrier and its flow rate was adjusted to 1.2ml/min. The analytical column connected to the system was an RTx-5 capillary column. The column head pressure was adjusted to 100 Kpa. Column temperature programmed from 40°C. The injector temperature was set at 230°C. The mass Spectra were screened range of M/Z 40-600amu.The relative percentage amount of each component was calculated by comparing its average peak area to the total areas [8, 9].

Identification of Components: Interpretation of mass spectrum GC-MS was conducted using the database of National Institute Standard and Technique (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The Compound Name, Molecular weight of the test material was ascertained [10].

RESULTS AND DISCUSSION: Plants contain many compounds with different structures. Extraction of these components depends on multiple factors and the most important of which are the solvent and the extraction method. Selection of the solvent and the extraction method depend on the different parts of the plant and their constituent materials. It is very difficult to select a certain solvent for each class of plant compounds because there are other substances along with these compounds that affect the solubility of the compounds of interest [11]. It should be noted that a high extraction yield for an extract does not necessarily mean a high content of active ingredients. According to the literature, despite the higher yield of an extraction method than another one, the contents of the extracted active ingredients such as phenolic compounds may be lower in the extract with the higher yield [12]. In lieu of this we have selected methanol as a solvent for the extraction of components from *C. dactylon*. A large number of components (68) have been identified in methanolic extract of the selected plant.

GC-MS Analysis of Components: The extraction and analysis of plant material play an important role in the development and quality control of

herbal formulation. Hence, present study was aimed to find out the bioactive compounds present in the methanolic extract of *Cynodon dactylon* by using gas chromatography and Mass spectroscopy. The active compounds with their peak number concentration (Peak area %) and retention time (RT) were presented on **Fig. 1** and **Table 1** which showed the presence of 68 compounds in the methanolic extract of *Cynodon dactylon*.

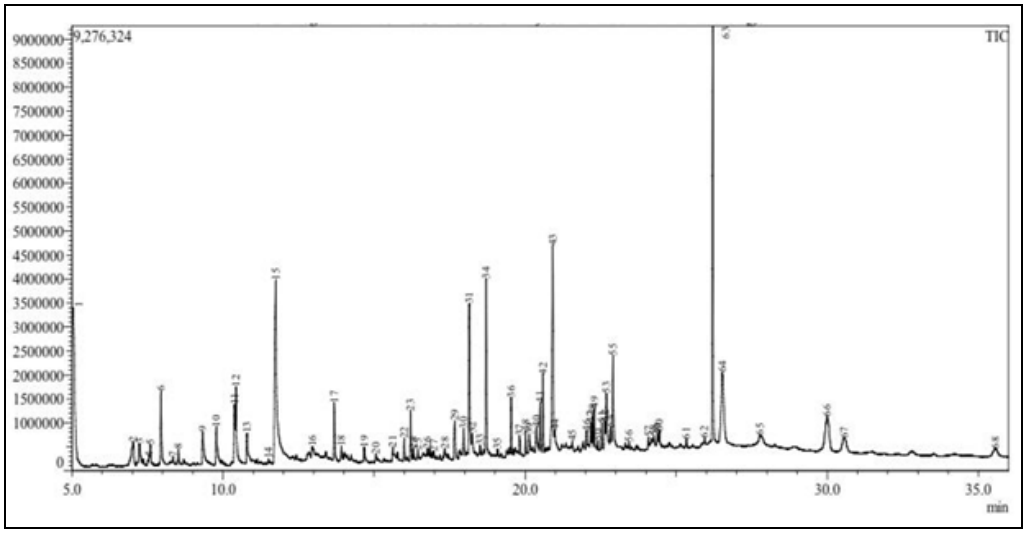

FIG. 1: GC-MS CHROMATOGRAM OF METHANOL EXTRACT OF *CYNODON DACTYLON* (L.) PERS

TABLE 1: GC-MS SPECTRAL ANALYSIS OF METHANOLIC EXTRACT OF *CYNODON DACTYLON* (L.) PERS

Peak#	Retention Time	Area%	Compound
1.	5.031	2.33	Furfural
2.	7.023	1.94	3-Aminopyrazine 1-oxide
3.	7.231	0.96	2-Furancarboxaldehyde,5-methyl
4.	7.550	0.18	Cyclotetrasiloxane,octamethyl
5.	7.596	0.48	2,4-Dihydroxy-2,5-dimethyl-3(2H)fura
6.	7.945	2.20	Oxazolidine,2,2-diethyl-3methyl
7.	8.351	0.20	1-Hexanol,2-ethyl-
8.	8.522	0.37	1,1carbonyldiimidazole
9.	9.326	1.40	Furayl hydroxymethyl ketone
10.	9.778	1.39	Levoglucosenone
11.	10.381	2.04	4H-Puran-4-one,2,3dihydro-3,5dihydroxy
12.	10.429	2.52	Hepta-2,4-dienoic acid methyl ester
13.	10.791	0.92	2-Furancarboxaldehyde,5-(chloromethyl)
14.	11.506	0.15	Benzaldehyde,2,4dimethyl
15.	11.744	10.92	5-hydroxymethylfurfural
16.	12.958	0.80	Valeric acid,2,3-epoxy-3,4dimethyl
17.	13.682	1.71	2H-pyran-2-one,3-acetyl-4-hydroxy
18.	13.920	0.37	Benzoic acid,3-ethoxy-4-hydroxy
19.	14.670	0.46	4H-1,3-benzodioxin-4-one
20.	15.061	0.32	2-chlorobenzoic acid decyl ester
21.	15.624	0.26	2,4-di-tert-butylphenol
22.	15.997	0.55	4-formyl-2,5-dimethoxy-6-methyltropo
23.	16.210	1.32	4-formyl-2,5-dimethoxy-6-methyltropo
24.	16.296	0.29	Dodecanoic acid
25.	16.450	0.23	2H-Pyran-5-carboxylic acid,4,6 dimeth
26.	16.765	0.24	Thiophene.2-butyl-5-hexyl
27.	16.973	0.25	1,3-dimethyl-1,3di(but-3-enyl)1,3-disi
28.	17.340	0.31	3-furancarboxylic acid,5-(ethoxymethyl)
29.	17.674	1.63	Tridecanoic acid,2-ethyl-2-methyl
30.	17.962	0.76	2-hydrazine-5,6,7,8-tetrahydro-3H-ben
31.	18.148	5.22	1H-Pyrole-2-carboxylic acid,4,5-dimethyl
32.	18.249	0.77	4H-1,3Benzodioxin-4-one,2-(1,1-dimethyl)

33.	18.506	0.34	Dodecyl cis-9,10-epoxyoctadecanoate
34.	18.708	4.70	3,5-diethoxycarbonyl-2,6-dimethylpyrine
35	19.085	0.20	1H-Pyrrole-2,5-dicarboxylic acid,3-methyl
36.	19.538	1.30	neophytadiene
37.	19.810	0.61	3,7,11,15-tetramethyl-2-hexadecen-1-0
38.	20.014	0.54	3,7,11,15-tetramethyl-2-hexadecen-1-0
39.	20.133	0.57	1H-2Benzopyran-3-one,7-ethoxy-4-hy
40.	20.366	0.99	1H-2Benzopyran-3-one,7-ethoxy-4-hy
41.	20.491	1.39	Hexadecanoicacid,methyl ester
42.	20.586	1.91	Benzenepropanoic acid,3,5-bis(1,1-dimethyl)
43.	20.913	7.05	n-Hexdecanoic acid
44.	20.983	0.39	Anthranilic acid,N-(2-Carboxy phenyl me
45.	21.562	0.37	Methyl2-ethylhexyl phthalate
46.	22.029	0.37	Phenanthro(3,2-b)furan-7/11-dione,1,2
47.	22.171	0.60	17-methoxy-d-homo-18-norandrosta-4
48.	22.226	0.72	9,12-octadecadienoic acid methyl ester
49.	22.287	0.92	9-octadecadirnoic acid methyl ester
50.	22.409	0.24	Acetic acid,3,7,11,15-tetramethyl-hexa
51.	22.534	0.61	Methyl stearate
52.	22.629	0.84	9,12-octadecadienoic acid
53.	22.687	2.27	9-octadecadienoic acid
54.	22.828	0.54	2-propenoic acid,3-(7-methoxy-2-oxo)
55.	22.906	2.95	Octadecanoic acid
56.	23.440	0.18	1-(2,3,4-trimethoxyphenyl)ethanol
57.	24.083	0.28	Terephthalic acid,piperidide,butylester
58.	24.256	0.24	9,9-diethoxy-7-nonyn-6-ol
59.	24.374	0.35	Cis-1-(4-isopropylphenyl)-3-(2-furyl)
60.	24.438	0.48	Fumaricacid,4chlorophenyl isohexyl
61.	25.334	0.27	Pyrazophos
62.	25.936	0.14	13-docosenoic acid,methyl ester
63.	26.209	11.71	Bis(2-ethylhexyl)phthalate
64.	26.522	5.43	tetrapentacontane
65.	27.772	1.39	Triacontanoic acid,methyl ester
66.	29.990	2.47	Phenol, 2,4-bis(1,1 dimethyl ethyl)
67.	30.564	2.01	tetrapentacontane
68.	35.565	1-13	tetrapentacontane

The prevailing components were levoglucosenone (R_t-9.778); Dodecanoic acid (R_t-16.296); Thiophene (R_t-16.765); Hexadecanoic acid (R_t-0.913) Neophytadiene (R_t-19.588); 9,12-Octadecadienoic acid (R_t-22.629); 2-Propenoic acid (R-$_t$22.828); terephthalic acid (R_t-24.083);Phenol, 2,4- bis (1, 1 dimethylethyl) (R_t-29.990).

Levoglucosenone is a chiral synthon for the synthesis of a variety of new and valuable compounds. n-Hexadecanoic acid has antioxidant, hypocholesterolemic nematicide, pesticide, lubricant, antiandrogenic, flavoring agent, Hemolytic and 5-Alpha reductase inhibitor. Octadecadienoic acid is known to show anti-inflammatory, hypocholesterolemic, cancer preventive, hepatoprotective, nematicide and Insectifuge, antihistaminic antieczemic, antiacne, 5-Alpha reductase inhibitor, antiandrogenic, antiarthritic and anticoronary agent. Dodecanoic acid has a role as a plant metabolite, an antibacterial agent and an algal metabolite. Thiophene are remarkably effective compounds both with respect to their biological and physiological functions such as anti-inflammatory, anti-psychotic, anti-arrhythmic, anti-anxiety, anti-fungal, antioxidant, estrogen receptor modulating, anti-mitotic, anti-microbial, kinases inhibiting and anti-cancer. Neophytadiene has a role as an anti-inflammatory agent, an antimicrobial agent, a plant metabolite and an algal metabolite.

CONCLUSION: The demand in the study of plants, which is one of the richest sources of promising versatile chemical compounds, is growing persistently throughout the world during the last few decades. Therefore, the data generated from these experiments provide the chemical basis for the wide use of this plant as therapeutic agent for treating various ailments. GC-MS method is a

direct and fast analytical approach for identification of phytoconstituents. The importance of the study is due to the biological activity of some of these compounds. The present study which revealed the presence of components in *C. dactylon* suggested that the contribution of these compounds on the pharmacological activity found helpful in knowing the particular action of this medicinal grass and can prove its medicinal importance as well as presence of valuable phytoconstituents in it applicable to various diseases and this fact revealing the wisdom of our ancestors and Rishis even in ancient times as they mentioned this grass in ancient literature.

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