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GC- MS Analysis of Spent Mushroom Compost (SMC) of oyster mushroom (*Pleurotus ostreatus)*

B* Chandrakala and Y. Sunila Kumari

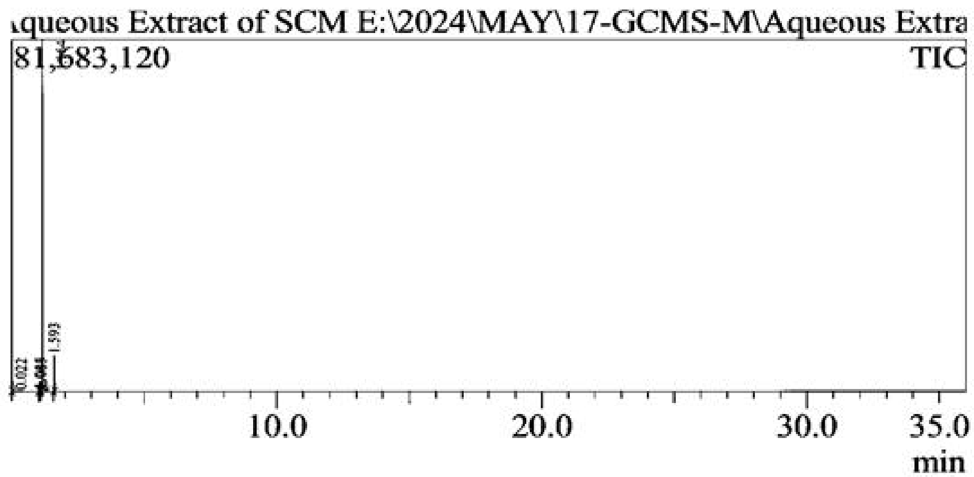
Department of Zoology, Osmania University College for Women (OUCW)
Hyderabad - 500095 (India)
*Corresponding author email: machachandrakala@gmail.com

Abstract

Gas Chromatography-Mass Spectrometry (GC-MS) is a robust technique for accurately identifying and quantifying the chemical constituents in a given sample. In the present study, aqueous extract of spent mushroom compost (SMC) was subjected to GC-MS analysis to identify its important entities, which can have anthelminthic properties. Our analysis revealed the presence of key element known as Propane, 1, 2-dimethoxy-. whose retention time (RT) was found to be 1. 164.minutes. This is the most prominent peak in the chromatogram, followed by Cyclopropyl carbinol, Diethylpropion Hydrochloride, nitrous oxide etc. From the chromatogram, it was quite evident that Propane, 1, 2-dimethoxy- was the main ingredient which may have anthelminthic properties. This compound is a potent dehydrating agent known to cause membrane damage by increasing reactive oxygen species (ROS) levels, ultimately destroying the bacteria. A similar mechanism may underlie its potential anthelmintic properties, which will be investigated in our future study upon Meloidogyne species which is the root knot nematode, infecting various plant species.

Key words : GC-MS analysis, spent mushroom compost (SMC), *Meloidogyne,* Propane, 1, 2-dimethoxy, reactive oxygen species (ROS), anthelminthic.

Root knot nematode, (*Meloidogyne* species) is a Phyto nematode, which causes root galls and swellings on the roots of the plants, frequently infecting quite a number of crops, such as tomato,[6], French bean[2], Mung bean[14], Winged bean (Simon, S., & Anamika.[12] *etc*. This infection causes significant damage to crops, leading to substantial economic losses in agriculture. In this context, when literature reveals, that spent mushroom compost when added to soil, showed considerable, decline in the infection of root knot nematode. Authors like Zhong-Yan Yang *et. al.,*[15] have reported that when spent mushroom compost synthesized using oyster mushroom, revealed that there was a 61% efficiency in control on nematode (*Meloidogyne incongnito*) infection on tobacco plants. Apart from this, studies by



Peak Report TIC

Pea	R.Time	Area	Area%	Height	A/H	Base m/z	Name
1	0.022	126797	0.15	217010	0.58	44.05	Cyclopropyl carbinol
2	1.044	258916	0.31	287365	0.90	44.05	Nitrous Oxide
3	1.083	602563	0.71	413992	1.46	44.05	Diethylpropion Hydrochloride
4	1.164	77401795	91.60	81592453	0.95	57.85	Propanc, 1,2-dimcthoxy-
5	1.593	6108018	7.23	8406660	0.73	43.10	Propane, 2,2-dimethoxy-
		84498089	100.00	90917480			

Fig-1. Chromatogram of aqueous extract of spent mushroom compost (SMC)

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Fig-2. Prominent peaks, with RT values

authors like Kharapude Chandrakant *et. al.,*[8] have used spent mushroom compost in combination with neem compost and successful in controlling *Meloidogyne* infection in Okra plants. A similar kind of observation has been reported by authors such as Bakr *et. al.,*[3], who have also employed spent mushroom compost to control root knot nematode in egg plant. Based upon these studies, it was quite evident that spent mushroom compost synthesized using oyster mushroom, seems to be a viable and eco-friendly option to contain the infection *Meloidogyne* infections. This also suggests that spent mushroom compost seems to contain, certain chemical constituents, which have an anthelminthic properties, and hence capable of controlling the nematode infections. In this regard, GC- MS (Gas chromatography, Mass Spec) analysis can reveal the presence of the principle constituents, which may be responsible for the specific properties of spent mushroom compost. However not many studies have been reported about the GC-MS analysis of spent mushroom compost. There are few reports, where in alcohol based extract of spent mushroom compost was analysed using GC-MS[7,10]. The present study aims to analyze the aqueous extract of oyster spent mushroom compost for the presence of key components that support its anthelmintic properties. Our objective is to evaluate the efficacy of spent mushroom compost as a potential anthelmintic against *Meloidogyne* under both *in vitro* and *in vivo* conditions. This analysis serves as a preliminary step to identify its most important and abundant constituents.

Preparation of Spent Mushroom compost (SMC) :

Spent mushroom substrate refers to residual waste generated in mushroom cultivation industry, typically containing a mixture of agricultural wastes such straw, animal manure, and inorganic substances *i.e.*, gypsum and limestone. Mushroom fruiting bodies belonging to oyster mushrooms (*Pleurotus ostreatus)* were obtained from the process of solid substrate fermentation while the submerged liquid fermentation was utilized for liquid spawn production used for mushroom cultivation. After the preparation, the compost was subjected to GC-MS analysis.

GC-MS Analysis of Spent mushroom compost (SMC) :

Aqueous SMC (Spent Mushroom compost) was analyzed by gas chromatography and mass spectrometry on a Shimadzu GC-MS-QP2020 NX equipped with an SH-Rxi-5Sil MS (30 x 0.25 x 0.25 mm) column at CMFT- RUSA-2.0 (O.U.) Hyderabad. The carrier helium gas was used at a rate of 1 ml/ min, and an injection volume of 1 L was used with an injector at 260°C and an ion source Temperature of 220°C. The temperature was kept constant at 50°C (isothermal for 4 minutes) with a 10°C/min increase up to 300°C and kept constant for 12 minutes. Metabolites' mass spectra were recorded at two scans per second with a scanning interval of 50-600 m/z. Metabolic compounds were detected using GC retention time and analyzed using standard mass spectral data from the Wiley and NIST (National Institute of Standards and Technology) Libraries 11.

In the present study, when spent mushroom compost (SMC) was subjected to GCMS, analysis, the chromatogram, showed different peaks, corresponding to different constituents. (Fig-1 and 2). The most prominent peak, was found to be that of Propane, 1, 2-dimethoxy-**.** whose RT was found to be 1.164.minutes. Other compounds include, Cyclopropyl carbinol, is a cyclo propyl derivative and seems to be an anti-tumor agent. Similarly a peak at 287365 corresponds to nitrous oxide. This compound is known to have antimicrobial properties. Similarly a peak at 413992 is identified to be that of a compound known as Diethylpropion Hydrochloride. This also is known to have antimicrobial properties and known to be mildly toxic. The abundance and importance of prominent peaks have been detailed as under.

In the present study, spent mushroom compost which was synthesized using oyster mushrooms was subjected to GC-MS analysis to learn about its chemical constituents. The chromatogram of the aqueous extract, exhibited quite a number of peaks, of which the prominent peak was found to be that of Propane, 1, 2-dimethoxy-**.**whose RT was found to be 1.164.minutes.This compound, is a known dehydrating agent, and can have beneficial effects when this compost is added to the soil. There are evidences, where in dehydrated compounds, or dehydrated fruit pulps or vegetable pulps are known to cause and increase in the antimicrobial properties[1,13]. This antimicrobial property may be attributed to its ability to devour the bacterial membranes, as suggested by authors, like Crowe, *et.al,*[4]; Prestrelski *et.al,*[9]; Schwager, *et.al,*[11]. These authors, opined that, dehydration process can damage the cell membranes by increasing the levels of reactive oxygen species (ROS), which can cause lipid peroxidation, denaturation of proteins and ultimately causing the death of cells. Based upon these evidences, it may be said that, the presence of Propane, 1, 2-dimethoxy in spent mushroom compost, which is known to be a potent dehydrating agent, can be an effective antimicrobial agent, and also probably be used as anthelminthic, against, *Meloidogyne* species (Root knot nematode), as it can cause an increase in ROS and thereby damage the cell membrane of the parasite leading to its extermination. The anthelminthic property of the spent mushroom compost will be tested for its potential anthelminthic activity in our *in vitro* and *in vivo* studies in future,

The GCMS analysis of spent mushroom compost, revealed the presence of Propane, 1, 2-dimethoxy compound as the primary constituent which has potent dehydrating capacity, which will be tested for its anthelminthic effects upon *Meloidogyne species* which is a root knot nematode, which frequently infects, several plant species causing huge damage to crops.

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