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Gas chromatography: Mass spectrometry (GC-MS) analysis of biomolecules from *Talaromyces pinophilus*

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Abstract
The aim of the study was to evaluate the biomolecule compounds present in *Talaromyces pinophilus* by Gas Chromatography – Mass Spectrometry (GC-MS). Fungi was isolated from the soil and molecularly identified as *Talaromyces pinophilus* which is found to be effective against root-knot nematode, *Meloidogyne incognita* under *in vitro* condition. Metabolomics analysis revealed the presence of major bioactive moetabolites such as Ethane, 1-chloro-2-nitro, Propanoic acid, 9-Octadecenoic acid methyl ester, n-Hexadecanoic acid, Oleic acid, 17-Pentatriacontene, Ethyl iso-allocholate, 7, 8-Epoxylanostan were identified and was reported to possess antinemic, antimicrobial, antifungal and antioxidants property.

Keywords: *Talaromyces pinophilus, Crude metabolites,* GC-MS, *Nematicidal property*

Introduction
The soilborne fungus *Penicillium pinophilum* (Synonymy *Talaromyces pinophilus*) has been demonstrated as a mycoparasitic fungus against *Botrytis cinerea* causing onion scalp and umbel blights. The genus *Talaromyces* was known as the fifth fungal genus for producing secondary metabolites (Zhai *et al.*, 2016) [10]. Bioactive secondary metabolites play a crucial role during the interactions within microbial communities, and may be related to different mechanisms, like competition for space/nutrients, parasitism and antagonism, as well as the induction of plant defence responses. Our research objective was to analyse the bioactive metabolites responsible for nematicidal activity by GC-MS analysis.

Materials and Methods
Fungal culture maintenance
The fungal culture, *Talaromyces pinophilus* was isolated from the soil and screened against root-knot nematode, *Meloidogyne incognita* under *in vitro* condition. Pure culture was maintained in potato dextrose agar medium and fungal disc was inoculated into the potato dextrose broth for culture filtrate preparation used for crude metabolites extraction.

Extraction of crude metabolites by solvent extraction method
Fungal disc was inoculated aspectically in the sterilized PDB and the flask was incubated @ 28 ± 2 ˚C for 15 days. After incubation, the culture filtrate was filtered through the Whatsman no. 1 filter paper and centrifuged the filtrate at 12000 rpm for 10 mins. Collected the suspension and discarded the pellets. Adjusted the pH of the filtrate to 2 by adding concentrated HCl. Solvent extract method was used to extract the crude metabolites and analysed by Gas Chromatography - Mass Spectrometry (GC-MS) at the Department of Nanoscience and Technology, TNAU, Coimbatore.

Results
Crude metabolites of *T. pinophilus* (fungal isolate F6) were extracted. Metabolites with antimicrobial, antioxidants, nematicidal and antifungal activity were identified as Ethane, 1-chloro-2-nitro, Propanoic acid, 9-Octadecenoic acid methyl ester, n-Hexadecanoic acid, Oleic acid, 17-Pentatriacontene, Ethyl Iso-allocholate, 7, 8-Epoxylanostan (Table 1) (Figure 1).

Discussion
Similar results were reported by Kumar *et al.* (2018) [4] that Hexadecanoic acid, methyl ester and 9-Octadecatrienoic acid has the potential nematicidal property.

Ethyl iso-allocholate was reported to exhibit anti-inflammatory, anticancer antimicrobial (Sarada *et al.*, 2012) [8]. Propanoic acid was isolated and identified from ethyl acetate which showed 50% juvenile mortality of *M. inocgnita* after 24 h inoculated at a concentration of 200µ/ml (Cao *et al.*, 2021) [2].

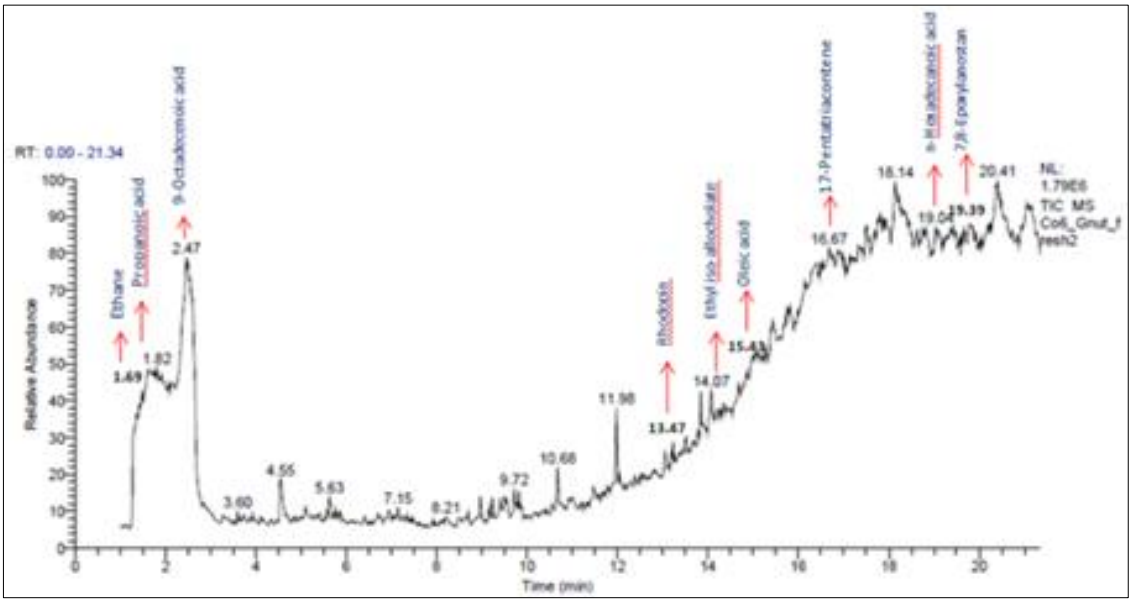

Fig 1: Chromatogram showing bioactive molecules of *Talaromyces pinophilus* by GC-MS chromatogram

Table 1: Characterization of secondary metabolites of *Talaromyces pinophilus*

S. No	Area (%)	RT	Compound name	Nature of compounds	Molecular formula	Molecular weight (g/mol)	Activity	References
1	28.42	1.69	Ethane, 1-chloro-2-nitro	Paraffin	$C_2H_4ClNO_2$	109.51	Anticancer Activity	Pravin *et al.* (2020) [7]
2	28.42	1.82	Propanoic acid	Saturated organic acid	$C_3H_6O_2$ (or) CH_3CH_2COOH	74.08	Nematicidial property	Abdel-Rahman *et al.* (2008) [1]
3	27.36	2.47	9-Octadecenoic acid methyl ester	Linoleic acid	$C_{18}H_{34}O_2$	282.5	Antimicrobial, Antiinflammatory, Hepatoprotective and Nematicide	George *et al.* (2018) [3]
4	2.56	16.67	17-Pentatriacontene	Alkanes	$C_{35}H_{70}$	490.9	Antiinflammatory Anticancer Antibacterial Antiarthritic	Kumar *et al.* (2018) [4]
5	1.78	14.07	Ethyl iso-allocholate	Steroid derivative	$C_{26}H_{44}O_5$	436.6	Antimicrobial, Antibacterial agent Antiinflammatory	Malathi and Ramaiah (2017) [5]
6	0.95	15.43	Oleic acid	Fatty acid	$C_{18}H_{34}O_2$ (or) $C_8H_{17}CH{=}CH(CH_2)_7COOH$	282.5	Nematicide, Antimicrobial property, Insectifuge, antiinflammatory, cancer preventive and hypocholesterolemic	Zhang *et al.* (2012) [11] Muitia *et al.* (2006) [6] Kumar *et al.* (2018) [4]
7	0.82	19.39	7,8-Epoxylanostan	Alcoholic compound	$C_{32}H_{54}O_4$	502.8	Anti-microbial property	Zekeya *et al.* (2014) [9]
8	0.43	19.04	n-Hexadecanoic acid	Palmitic acid methyl ester	$C_{21}H_{46}O_2Si_2$	386.8	Antioxidant, Hypocholesterolemic, Nematicide, Pesticide, Lubricant, Anti androgenic	Pravin *et al.* (2020) [7]

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