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GC-MS Analysis of Phytocomponents In the Ethanolic Leaf Extract of *Allium Fistulosum* Linn

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ABSTRACT

The present study investigates the phytochemical composition of the ethanolic leaf extract of *Allium fistulosum* L. (Welsh onion) using Gas Chromatography–Mass Spectrometry (GC–MS). The analysis identified 38 bioactive compounds, including fatty acids, esters, sterols, alcohols, phenolics, and other secondary metabolites. Major constituents detected were n-hexadecanoic acid (27.81%), DL-Proline, 5-oxo-, ethyl ester (10.77%), γ-sitosterol (7.94%), 9, 12, 15-octadecatrienoic acid (4.60%), and phytol (4.22%). These compounds are reported to exhibit various pharmacological activity such as antimicrobial, antioxidant, anti-inflammatory, and potential anticancer activities. The phytoconstituents identified highlight the therapeutic relevance of *A. fistulosum* and provide scientific support for its traditional medicinal applications. The findings further suggest potential utility of this plant in pharmaceutical, nutraceutical, and cosmetic formulations.

Keywords: GC-MS study, Therapeutic, Phytocomponents, Chromatogram

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INTRODUCTION

Medicinal plants are among the earliest forms of therapy, having been used in traditional medicine across different cultures worldwide for thousands of years. Over generations, communities have preserved and transmitted empirical knowledge of their therapeutic benefits. Many plant species have had their biological activities and bioactive constituents identified, while others remain underexplored or require further scientific investigation. Despite these gaps, medicinal plants continue to hold promising potential for future healthcare applications. [1]

The Allium genus has a long history of use in traditional medicine. Modern research on Allium species now focuses on evaluating the pharmacological properties of their extracts and active compounds. [2] Among them, green onions (*Allium fistulosum*) commonly referred to as scallion, Welsh onion, Japanese bunching onion, or spring onion belong to the Amaryllidaceae family and are well known for their distinctive flavour and wide culinary applications. [3]

Extracts derived from green onions have demonstrated various therapeutic potentials, including anti-inflammatory, antimicrobial, anticancer, and anti-arthritic effects. Their medicinal significance has been acknowledged for centuries, particularly within traditional healing practices. [4] Different plant parts such as bulbs, pseudostem juice, leaves, flowers, seeds, and roots are valued for multiple bioactivities, including antibacterial, antitumor, antihypertensive, anti-obesity, antioxidant, cardiovascular activation, antiplatelet aggregation, relief of intestinal spasms, and immune system regulation. The key bioactive constituents include volatile oils (mainly sulphides), oleic acid, linoleic acid, allicin, pectin, and vitamin C. [5] The present study aims to identify phytoconstituents in the ethanolic leaf extract of *Allium fistulosum* Linn. Using GC-MS analysis, to evaluate its medicinal potential.

MATERIALS AND METHOD

Plant Material Collection, Authentication

The leaves of *Allium fistulosum* l., were collected on 27 January 2025, from Thrissur vegetable market, district of Kerala. The plant specimen had been taxonomically identified and authenticated by Dr. Ranjusha A. P., Head of the Department of Botany at N. S. S. College, Ottapalam. The leaves were separated from the stem, thoroughly washed and they were shade-dried for about 25 to 30 days, ground into powder using a mixer grinder and kept in an airtight storage unit for subsequent studies

Preparations Of Plant Powder

The entire *Allium fistulosum* plant was washed and allowed to dry in the shade for a period of 25 to 30 days. After being ground in a mixer to a coarse powder, the dried plant was sealed in an airtight

container. In the phytochemical research, extraction is the first step. It involves applying certain solvents and following established protocols to separate the parts of plants that have therapeutic value.

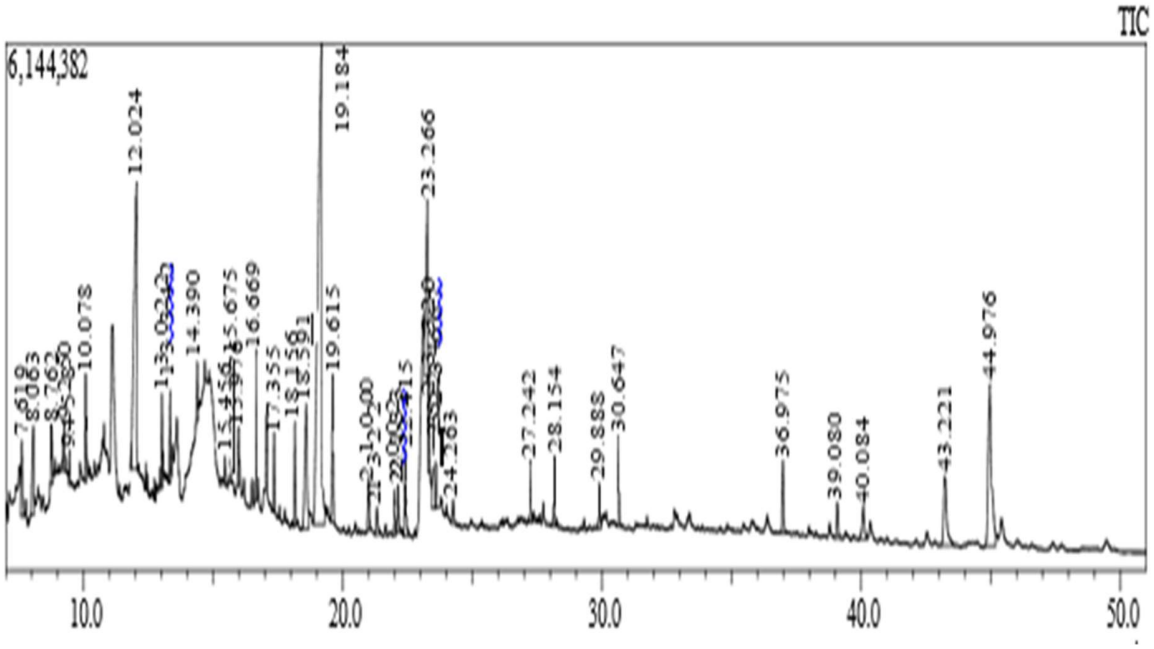
Preparation Of Extract

Using a Soxhlet apparatus, 25 g of the dried, coarsely ground plant material of *Allium fistulosum* L. was extracted with a high-polarity solvent (ethanol) at 60° to 70° for 18 hours, or until the solvent became colorless in the siphon tube. The extracts were concentrated using a rotary evaporator. The resulting semisolid residue was gathered and kept in desiccators. The extract was subjected to GC-MS analysis. [6]

Characterization by GC-MS

The ethanol extract of Allium fistulosum leaves was subjected to gas chromatography–mass spectrometry (GC–MS) analysis at the Kerala Forest Research Institute (KFRI) on 23rd July 2025 to identify its phytochemical constituents. The GC–MS procedure can be carried out under the following conditions. The sample should first be clarified by filtration or centrifugation, followed by extraction of analytes with an appropriate solvent such as hexane or dichloromethane. An internal standard may be added, and the extract concentrated if necessary, before transferring to a clean auto sampler vial. A capillary GC column (30 m × 0.25 mm × 0.25 µm, nonpolar phase) is used, with helium as the carrier gas at a constant flow rate of 1.0 mL/min. The injection port temperature is set between 250–280 °C, and depending on analyte concentration, either split or splitless injection is employed, with 0.5–1.0 µL sample volume. The oven is programmed to start at 40 °C (held for 2 min), increase at 10 °C/min to 150 °C, then 5 °C/min to 250 °C, and further at 10 °C/min to 300 °C, with a final hold of 5–10 min, ensuring the total run includes the 7.00–51.00 min MS acquisition range. A solvent delay of 6.50 min is applied to protect the detector from solvent peaks. In the MS system, the ion source temperature is maintained at 230 °C, the interface at 280 °C, with electron ionization at 70 eV and a detector gain of 0.90 kV relative to tuning. Data are collected in scan mode across m/z 35–500 with an event time of 0.30 s and a scan speed of 1666. The analysis begins with a blank run to monitor background, followed by calibration standards, quality control injections, and then the sample runs, with QC checks included at intervals. Peaks are identified by comparing spectra with reference libraries such as NIST or Wiley, confirmed against standards when available, and quantified using calibration curves with the internal standard. Throughout the procedure, good laboratory practices are followed, including the use of PPE, handling solvents under a fume hood, safe waste disposal, and troubleshooting for sensitivity, peak shape, or carryover issues when required.[7]

RESULTS AND DISCUSSION

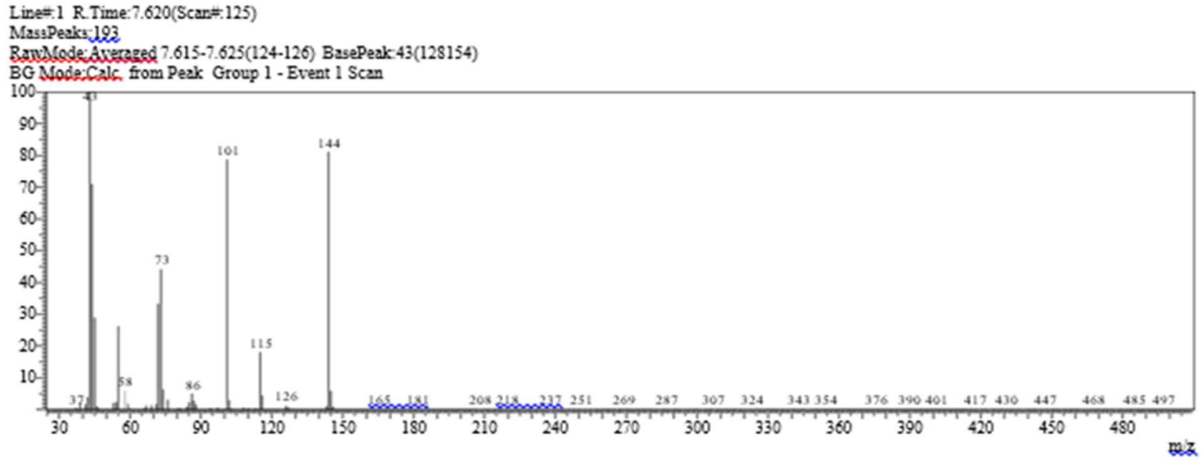


Graph 1: Chromatogram of EEAF

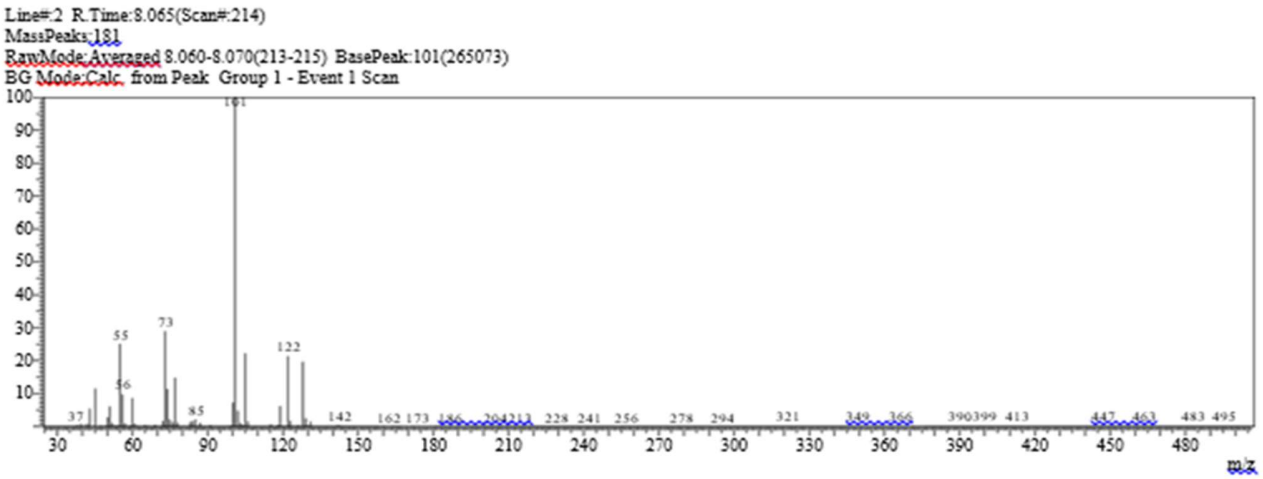
Table 1: GC-MS of EEAF

Peak	R. time	Mol wt	Formula	Compound Name
1	7.619	144	$C_6H_8O_4$	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-
2	8.063	146	$C_6H_{10}O_4$	Ethyl hydrogen succinate
3	8.762	120	C_8H_8O	4-Vinylphenol
4	9.250	136	$C_8H_8O_2$	Benzeneacetic acid
5	9.458	158	$C_9H_{18}O_2$	Nonanoic acid
6	10.078	150	$C_9H_{10}O_2$	2-Methoxy-4-vinylphenol
7	12.024	157	$C_7H_{11}NO_3$	DL-Proline, 5-oxo-, ethyl ester
8	13.022	168	$C_9H_{12}O_3$	Homovanillyl alcohol
9	13.342	200	$C_{12}H_{24}O_2$	Dodecanoic acid
10	14.390			No hit compound
11	15.456	226	$C_{13}H_{22}O_3$	7-Oxabicyclo[4.1.0] heptan-3-ol, 6-(3-hydroxy-1-butenyl)-1,5,5-trimethyl-
12	15.675	228	$C_{14}H_{28}O_2$	Tetradecanoic acid
13	15.976	196	$C_{11}H_{16}O_3$	6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one
14	16.669	278	$C_{20}H_{38}$	Neophytadiene
15	17.355	278	$C_{20}H_{38}$	Neophytadiene
16	18.156	270	$C_{17}H_{34}O_2$	Hexadecanoic acid, methyl ester
17	18.591	296	$C_{20}H_{40}O$	Phytol
18	19.184	256	$C_{16}H_{32}O_2$	n-Hexadecanoic acid
19	19.615	284	$C_{18}H_{36}O_2$	Hexadecanoic acid, ethyl ester
20	21.000	268	$C_{17}H_{32}O_2$	cis-10-Heptadecenoic acid
21	21.322	270	$C_{17}H_{34}O_2$	Heptadecanoic acid
22	22.002	294	$C_{19}H_{34}O_2$	9,12-Octadecadienoic acid (Z, Z)-, methyl ester
23	22.133	292	$C_{19}H_{32}O_2$	9,12,15-Octadecatrienoic acid, methyl ester, (Z, Z,Z)-

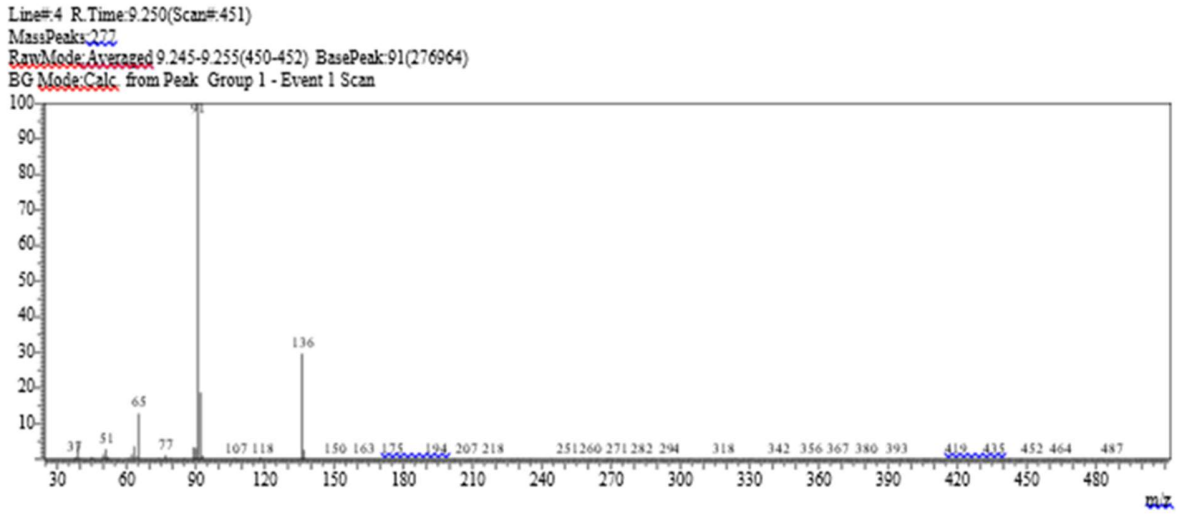
24	22.415	296	$C_{20}H_{40}O$	Phytol
25	23.266	278	$C_{18}H_{30}O_2$	9,12,15-Octadecatrienoic acid, (Z, Z,Z)-
26	23.320	310	$C_{20}H_{38}O_2$	cis-13-Eicosenoic acid
27	23.524	308	$C_{20}H_{36}O_2$	Linoleic acid ethyl ester
28	23.667	352	$C_{21}H_{36}O_4$	Linolenic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester (Z, Z,Z)-
29	24.263	312	$C_{20}H_{40}O_2$	Octadecanoic acid, ethyl ester
30	27.242	280	$C_{20}H_{40}$	Cyclohexane, tetradecyl-
31	28.154	382	$C_{26}H_{54}O$	1-Hexacosanol
32	29.888	274	$C_{16}H_{31}ClO$	Palmitoyl chloride
33	30.647	390	$C_{24}H_{38}O_4$	Bis(2-ethylhexyl) phthalate
34	36.975	402	$C_{27}H_{46}O_2$	delta.-Tocopherol
35	39.080	384	$C_{27}H_{44}O$	Cholesta-4,6-dien-3-ol, (3.beta.)-
36	40.084	386	$C_{27}H_{46}O$	Cholesterol
37	43.221	412	$C_{29}H_{48}O$	Stigmasterol
38	44.976	414	$C_{29}H_{50}O$	gamma. -Sitosterol



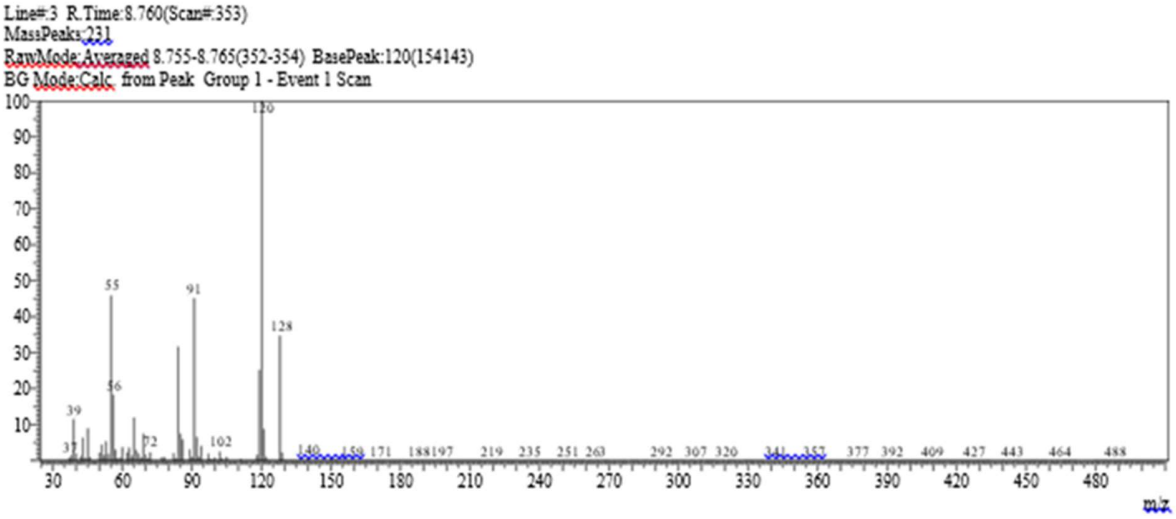
Graph 2: Chromatogram of 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-



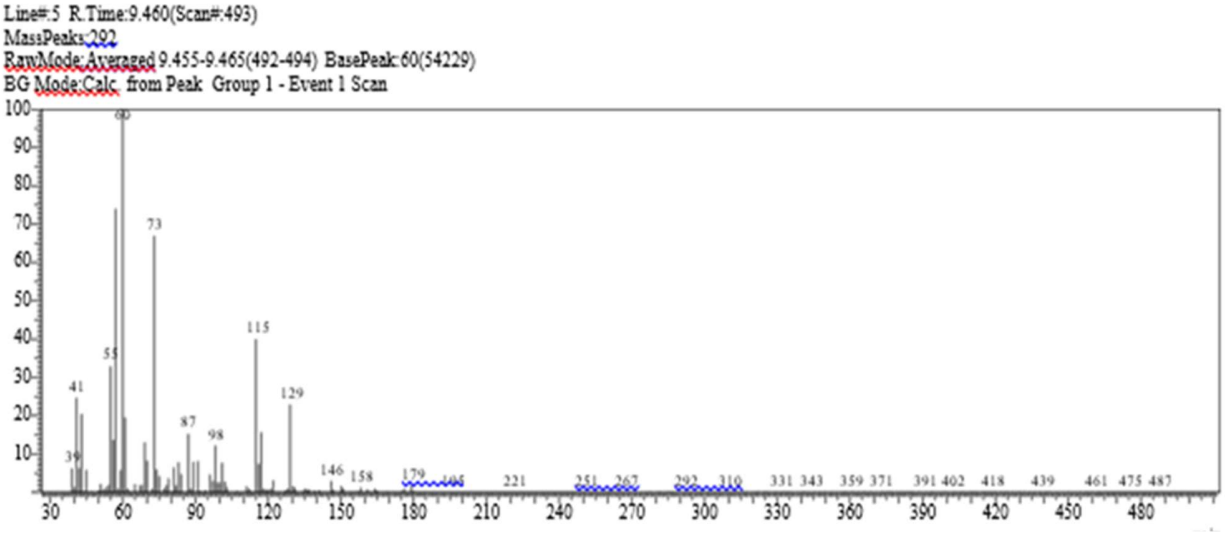
Graph 3: Chromatogram of Ethyl hydrogen succinate



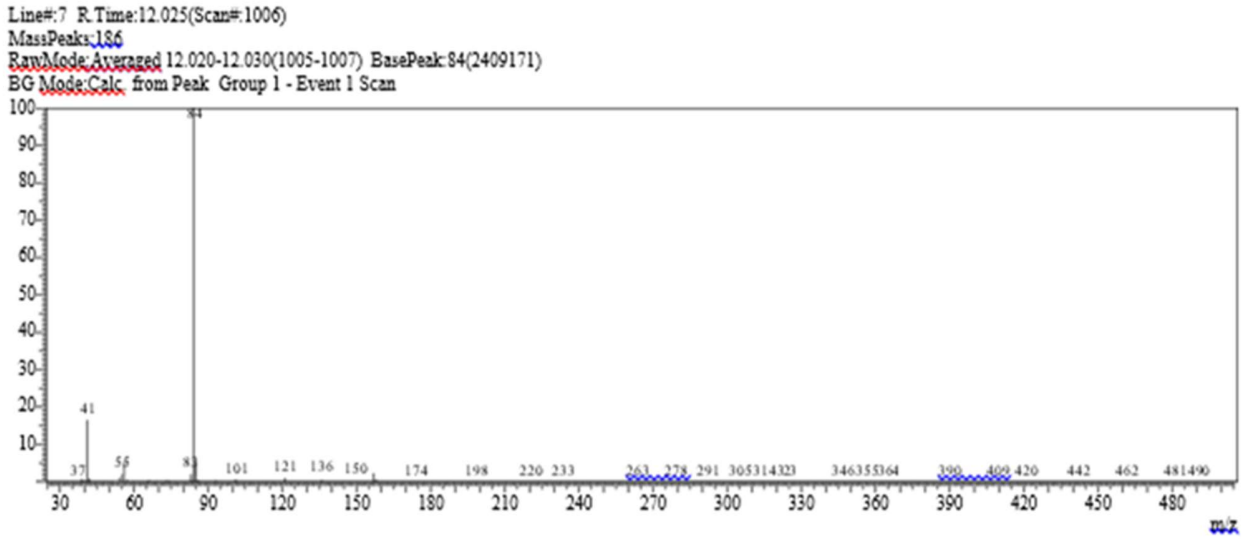
Graph 4: Chromatogram of 4-Vinylphenol



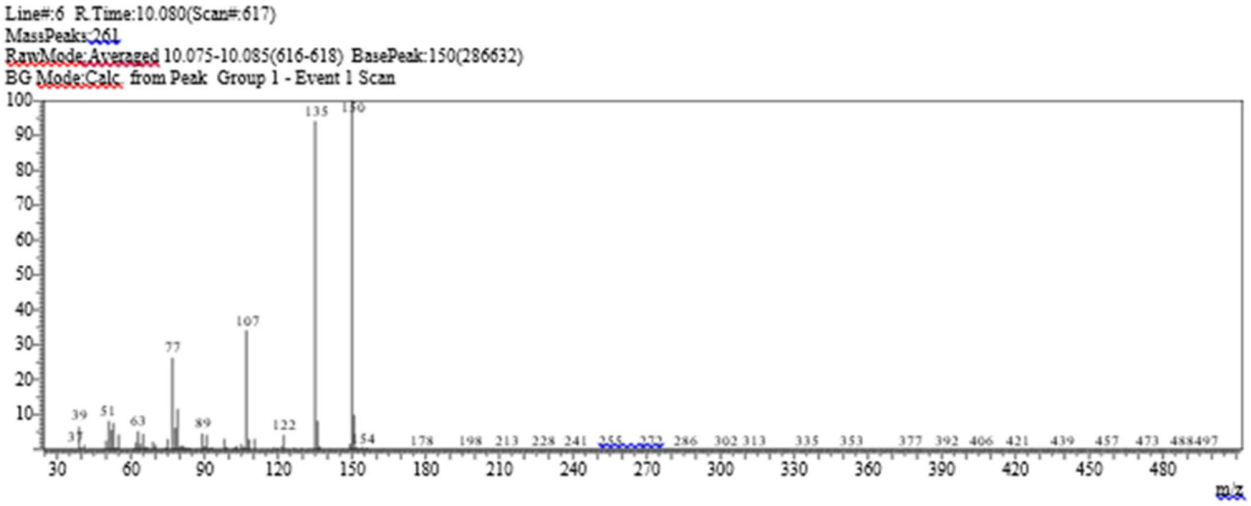
Graph 5: Chromatogram of Benzeneacetic acid



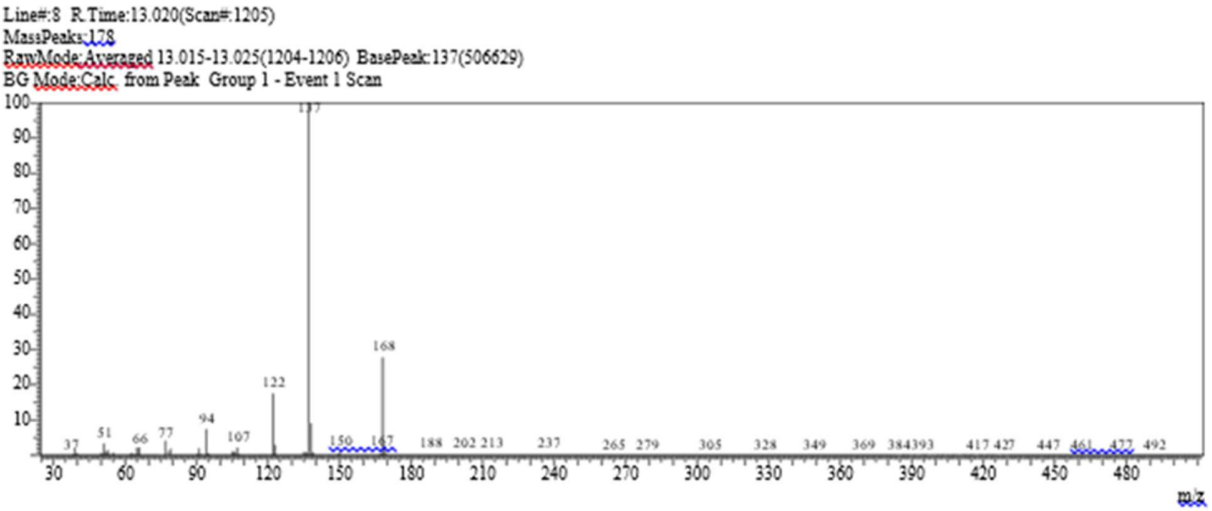
Graph 6: Chromatogram of Nonanoic acid



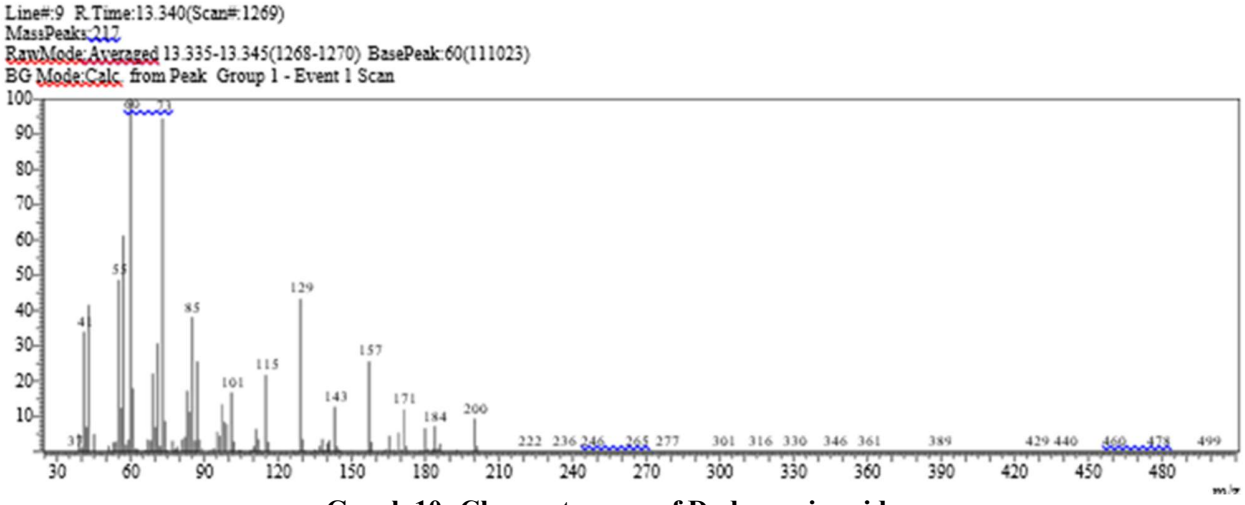
Graph 7: Chromatogram of 2-Methoxy-4-vinylphenol



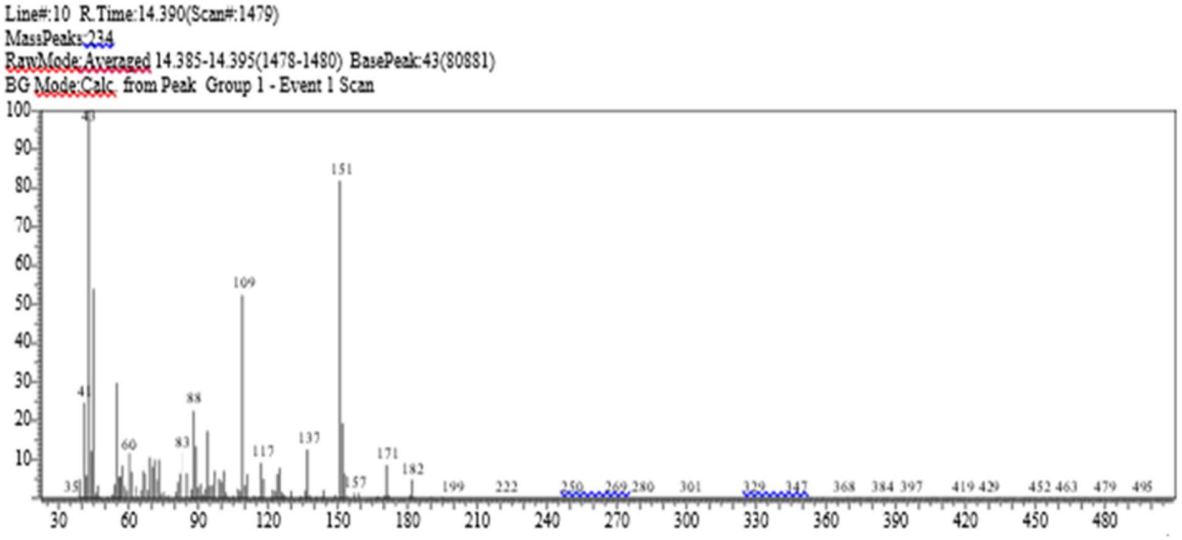
Graph 8: Chromatogram of DL-Proline, 5-oxo-, ethyl ester



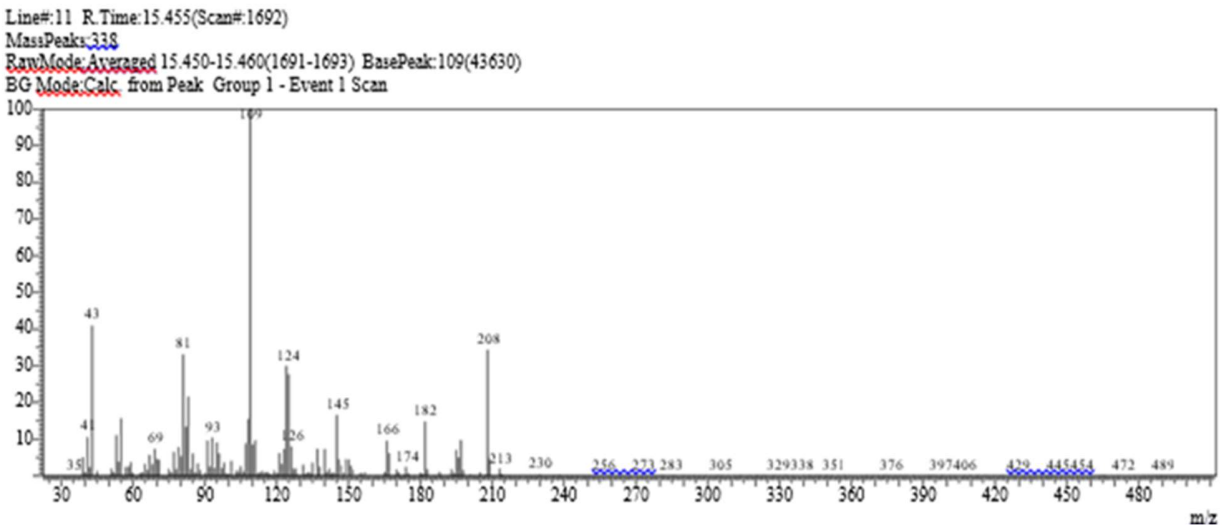
Graph 9: Chromatogram of Homovanillyl alcohol



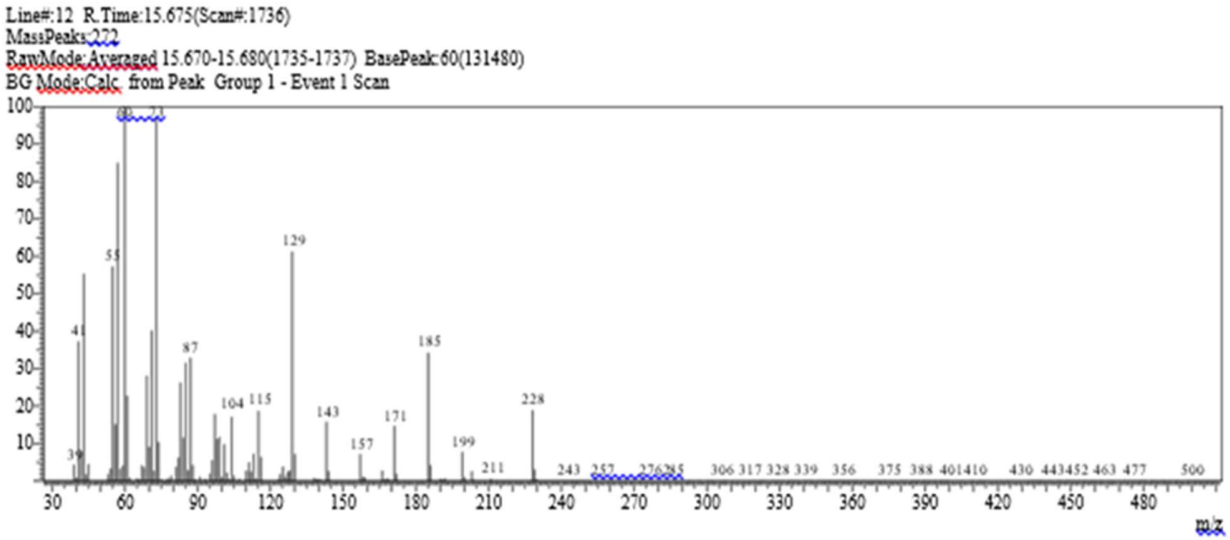
Graph 10: Chromatogram of Dodecanoic acid



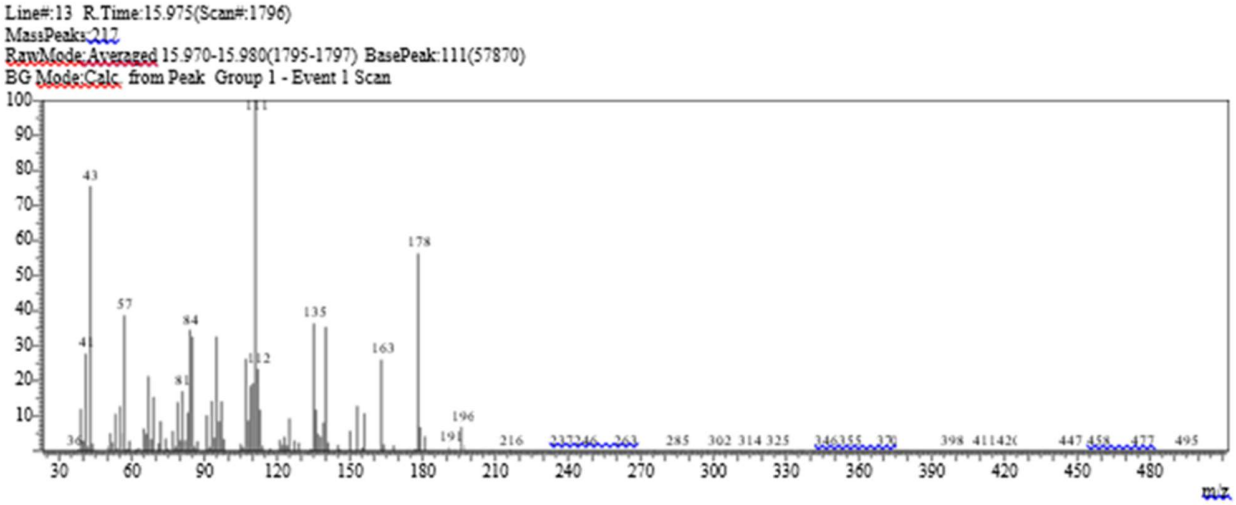
Graph 11: No hit compound



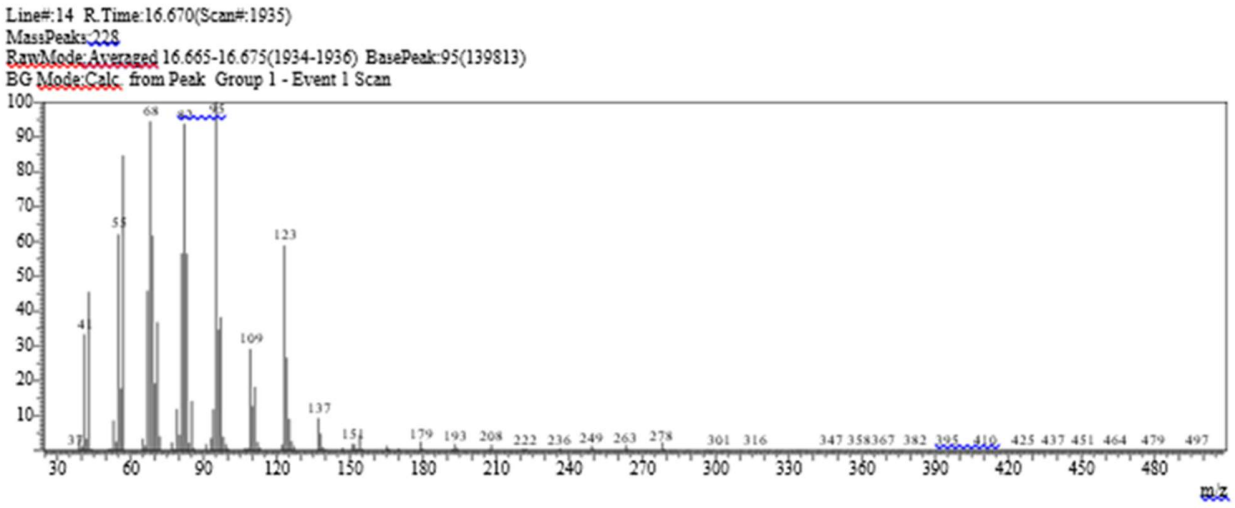
Graph 12: Chromatogram of 7-Oxabicyclo[4.1.0]heptan-3-ol, 6-(3-hydroxy-1-butenyl)-1,5,5-trimethyl



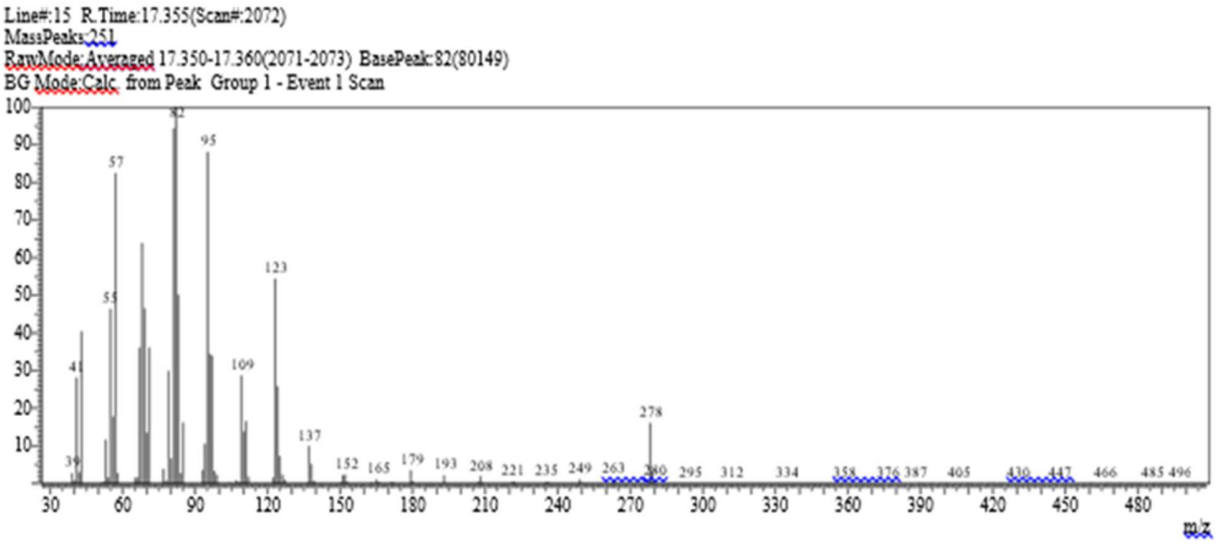
Graph 13: Chromatogram of Tetradecanoic acid



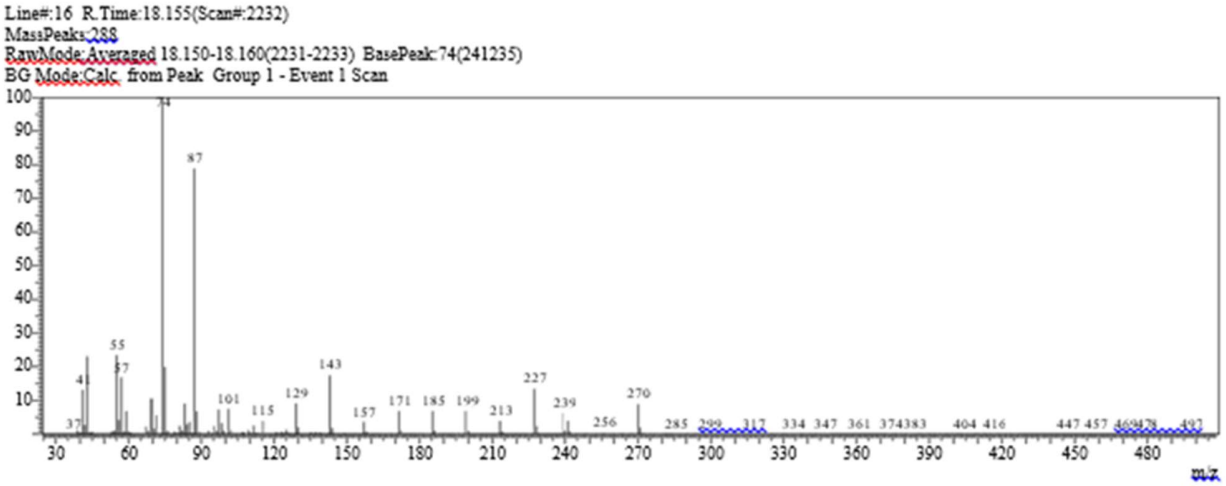
Graph 14: Chromatogram of 6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a- tetrahydrobenzofuran-2(4H)-one



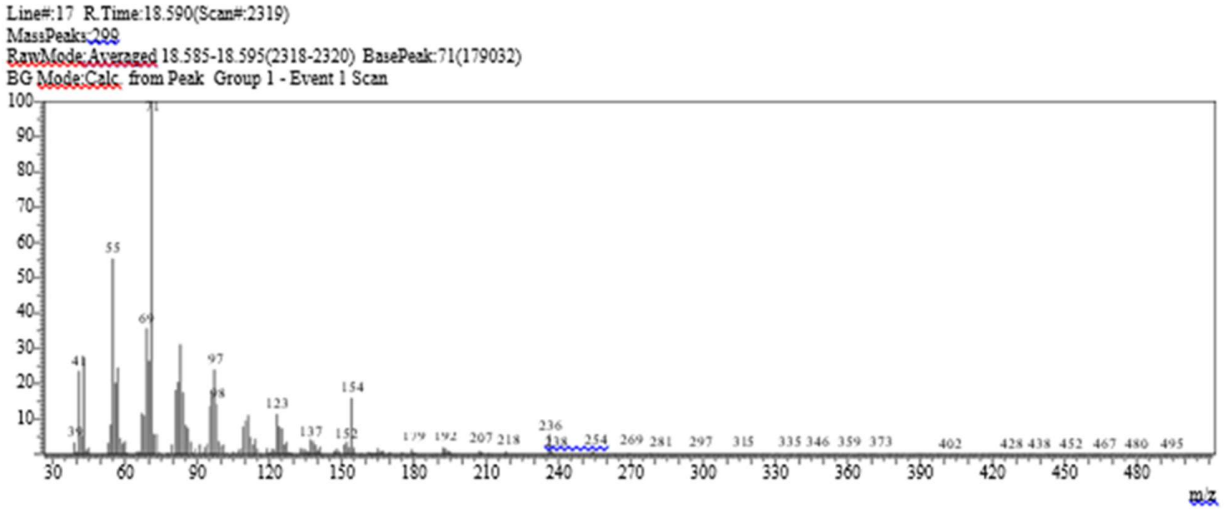
Graph 15: Chromatogram of Neophytadiene



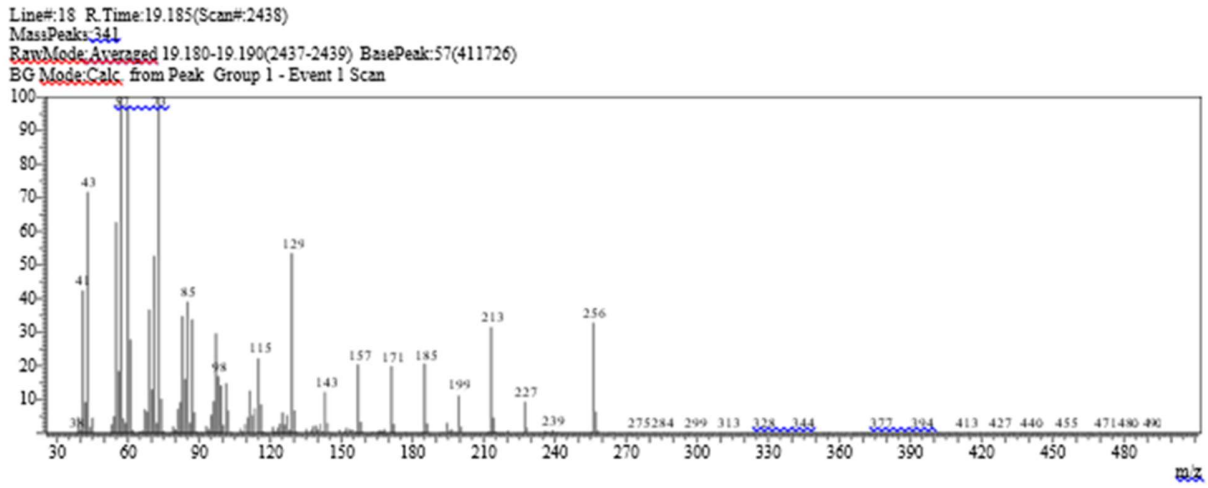
Graph 16: Chromatogram of Neophytadiene



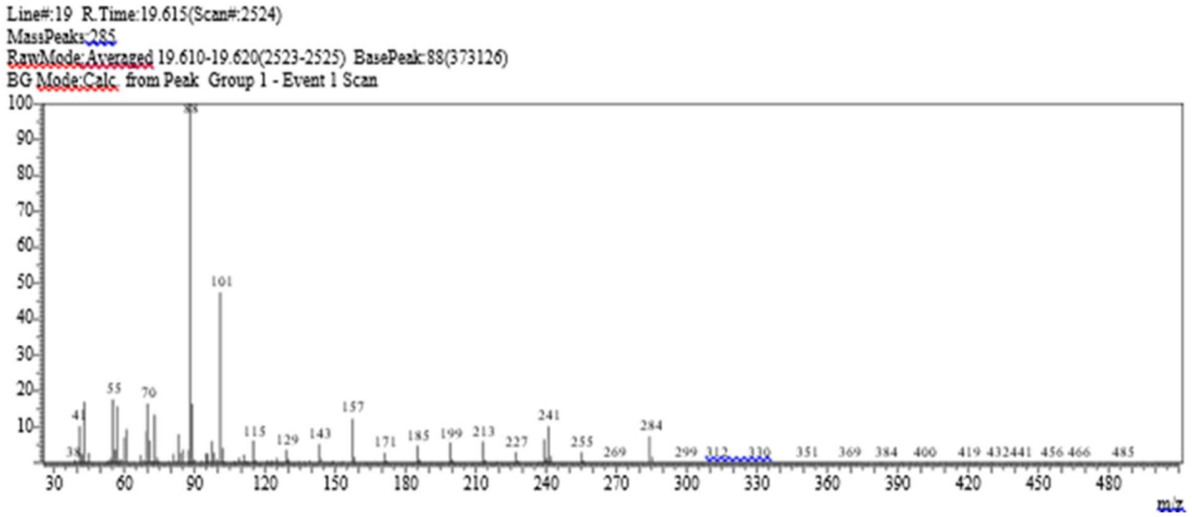
Graph 17: Chromatogram of Hexadecanoic acid, methyl ester



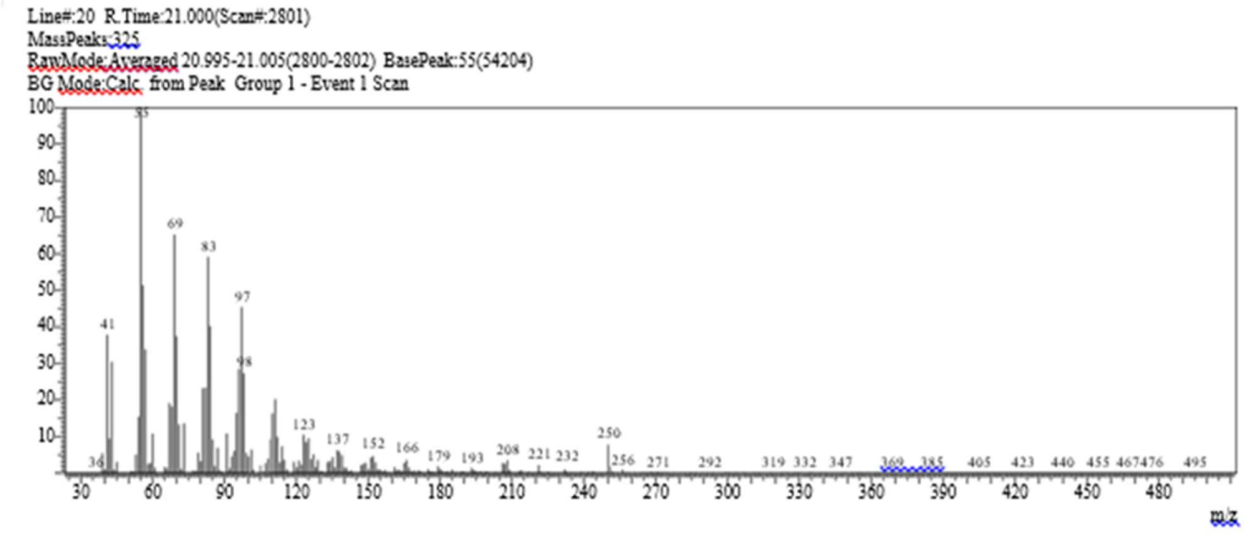
Graph 18: Chromatogram of Phytol



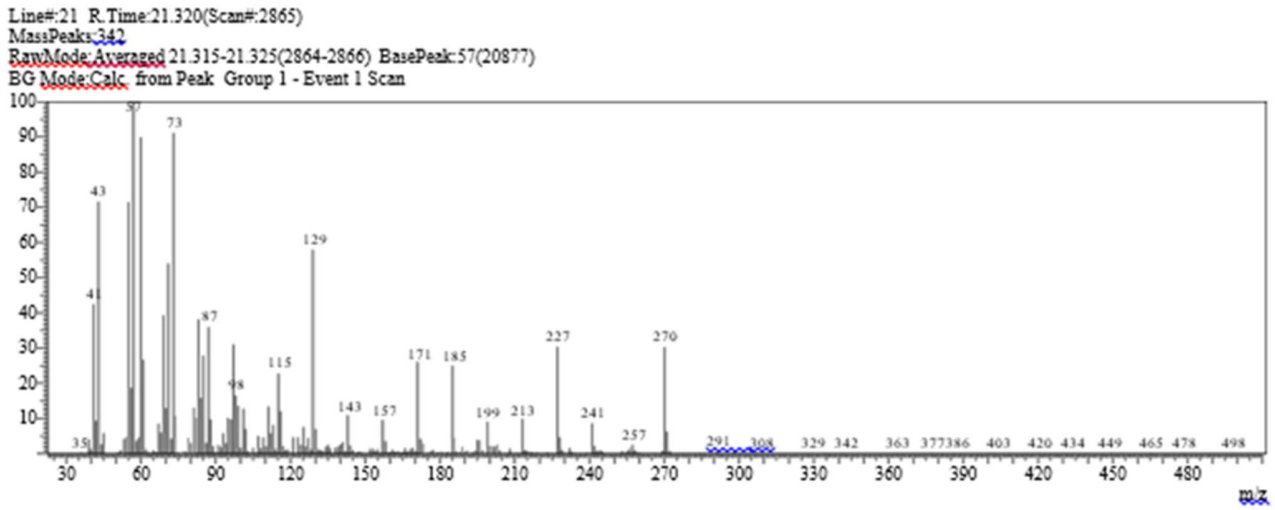
Graph 19: Chromatogram of n-Hexadecanoic acid



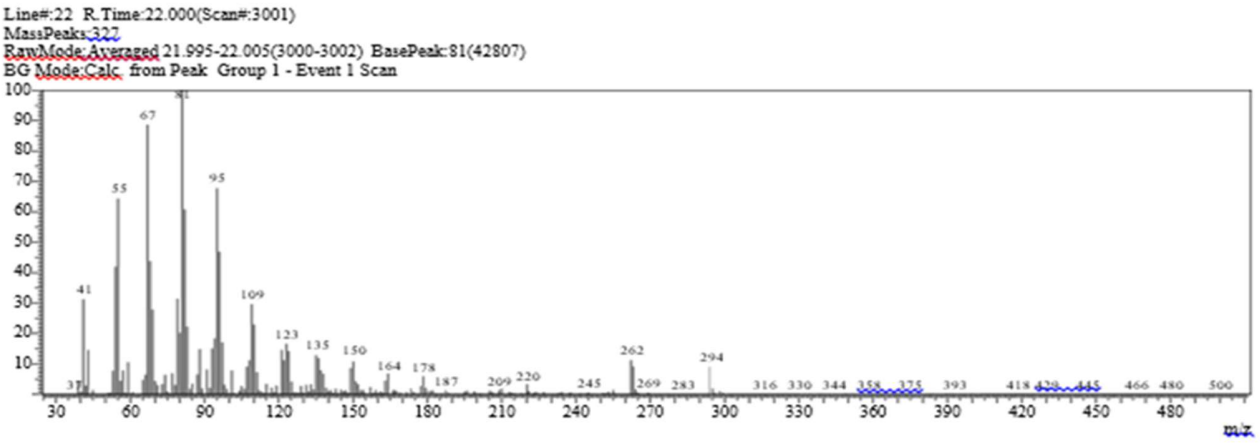
Graph 20: Chromatogram of Hexadecanoic acid, ethyl ester



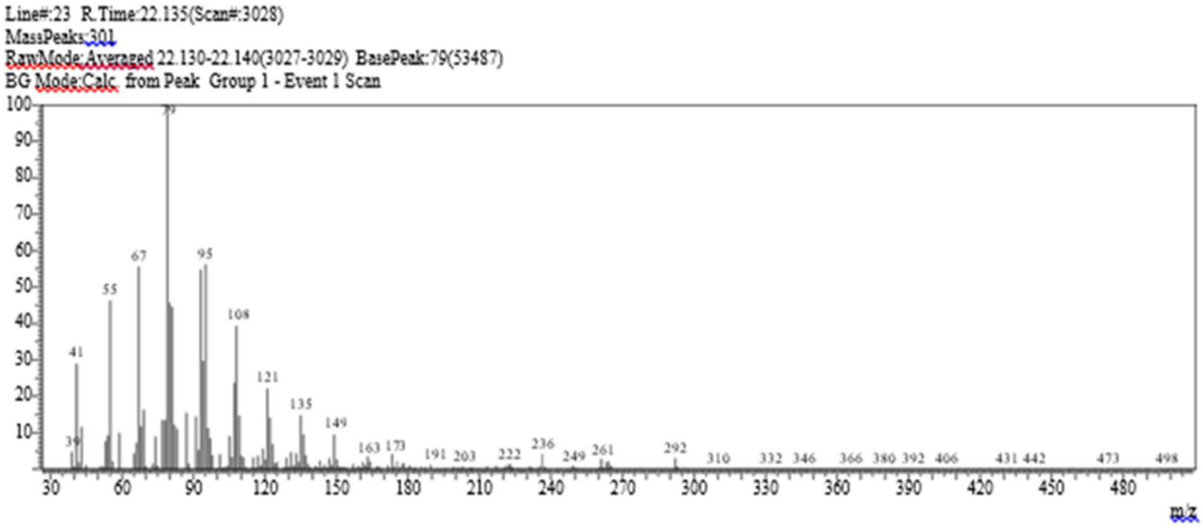
Graph 21: Chromatogram of cis-10-Heptadecenoic acid



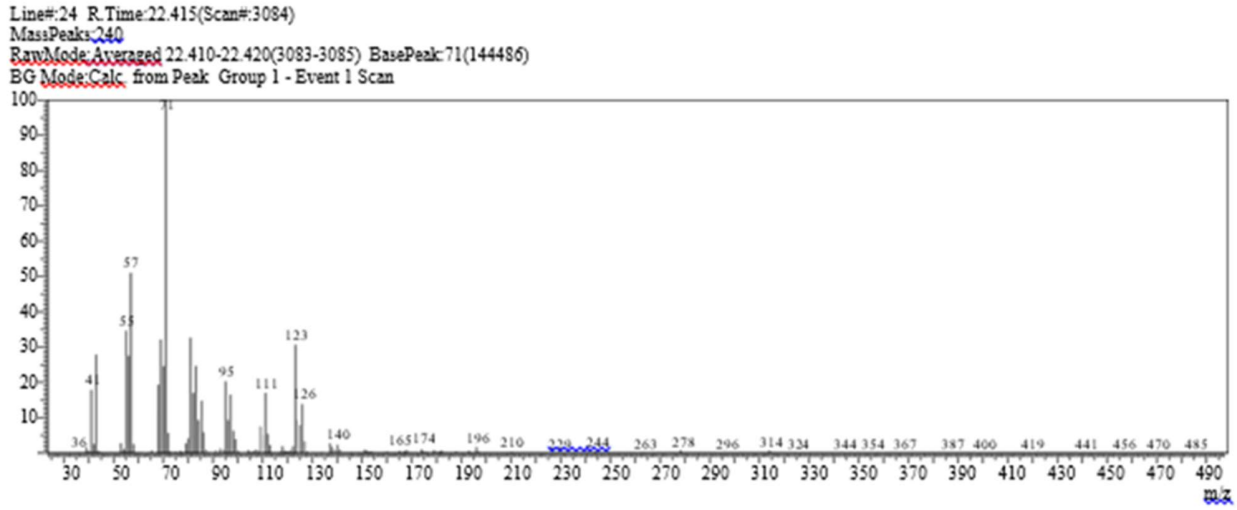
Graph 22: Chromatogram of Heptadecanoic acid



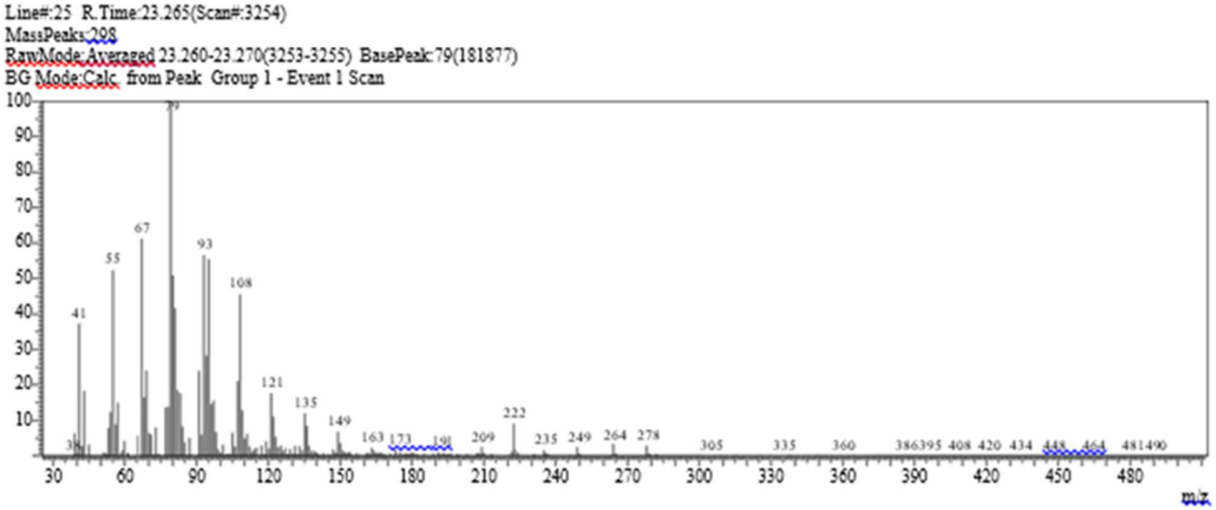
Graph 23: Chromatogram of 9, 12-Octadecadienoic acid (Z,Z)-, methyl ester



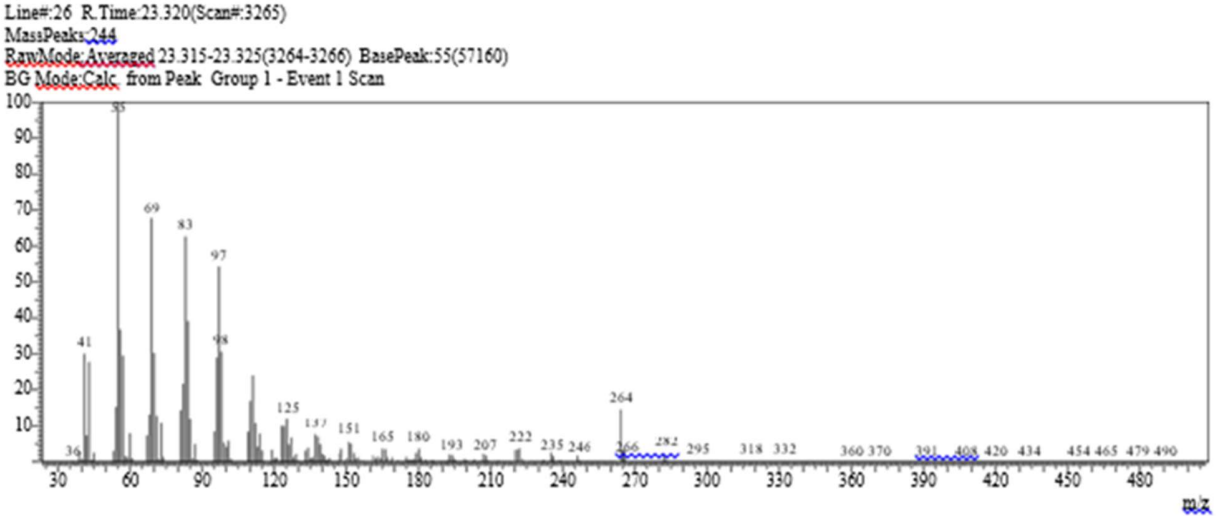
Graph 24: Chromatogram of 9, 12, 15-Octadecatrienoic acid, methyl ester, (Z, Z,Z)-



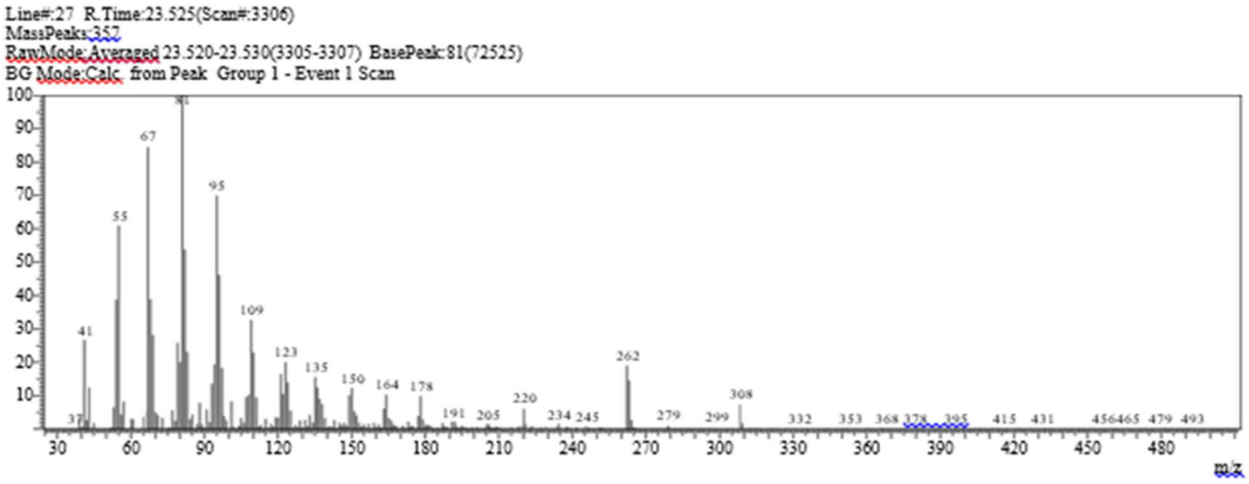
Graph 25: Chromatogram of phytol



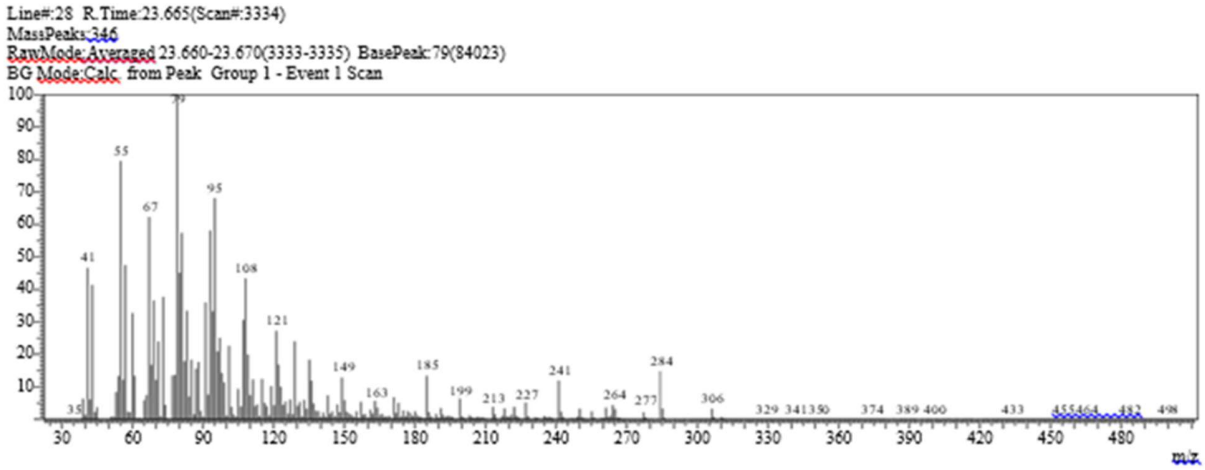
Graph 26: Chromatogram of 9, 12, 15-Octadecatrienoic acid, (Z,Z,Z)-



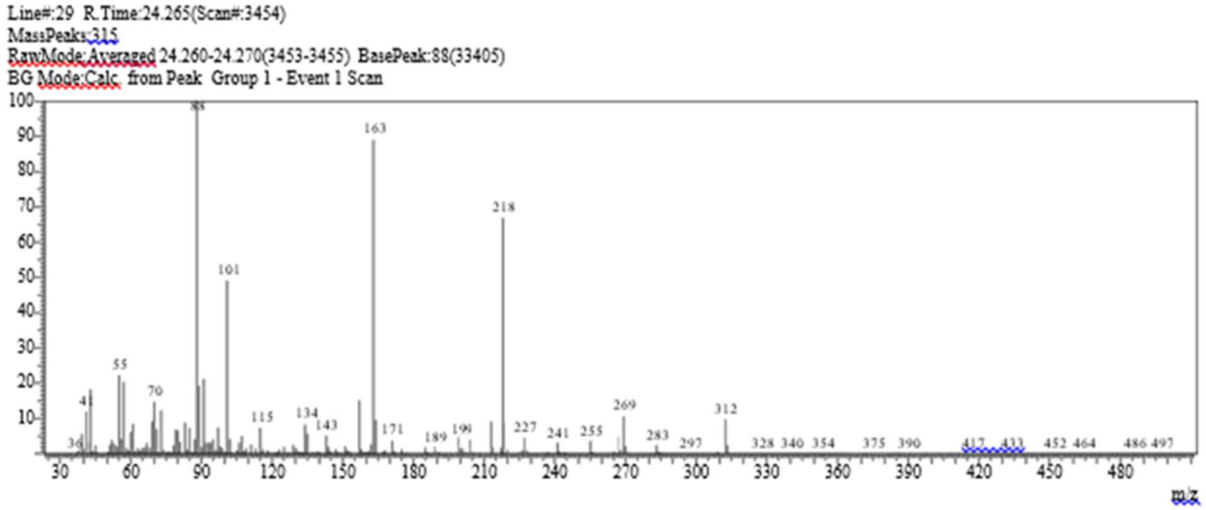
Graph 27: Chromatogram of cis-13-Eicosenoic acid



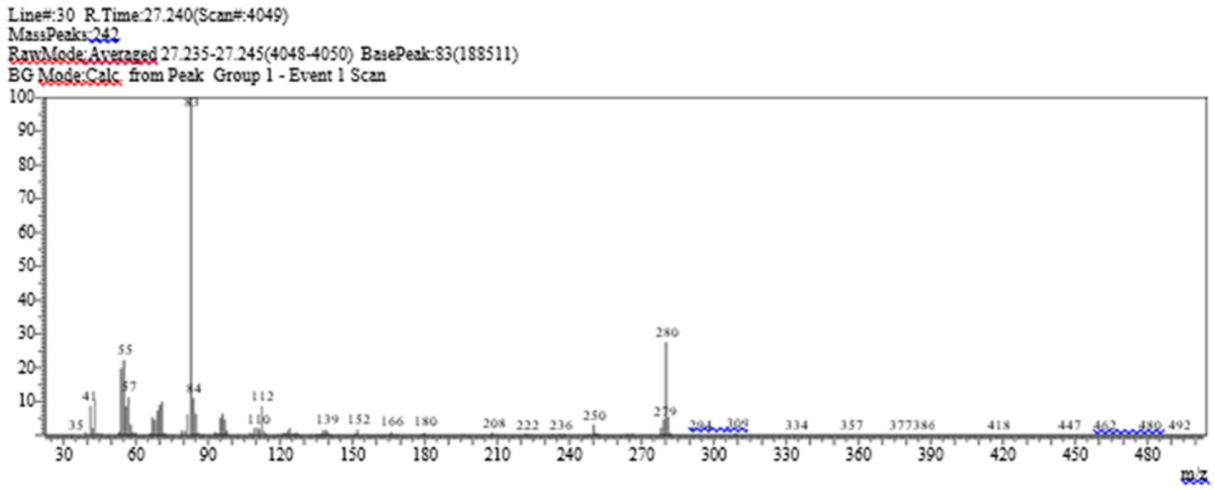
Graph 28: Chromatogram of Linoleic acid ethyl ester



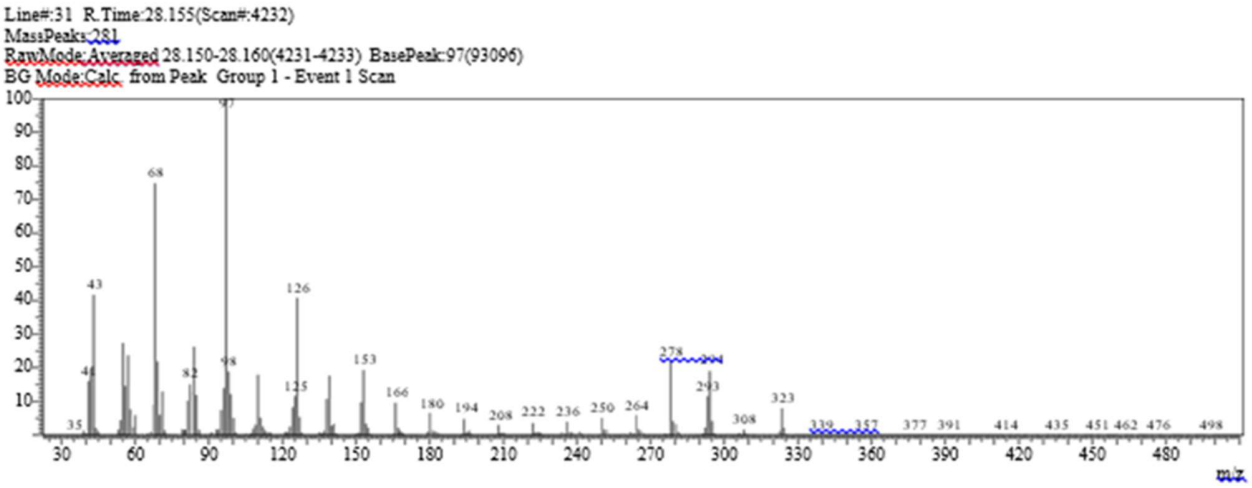
Graph 29: Chromatogram of Linolenic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester (Z,Z,Z)-



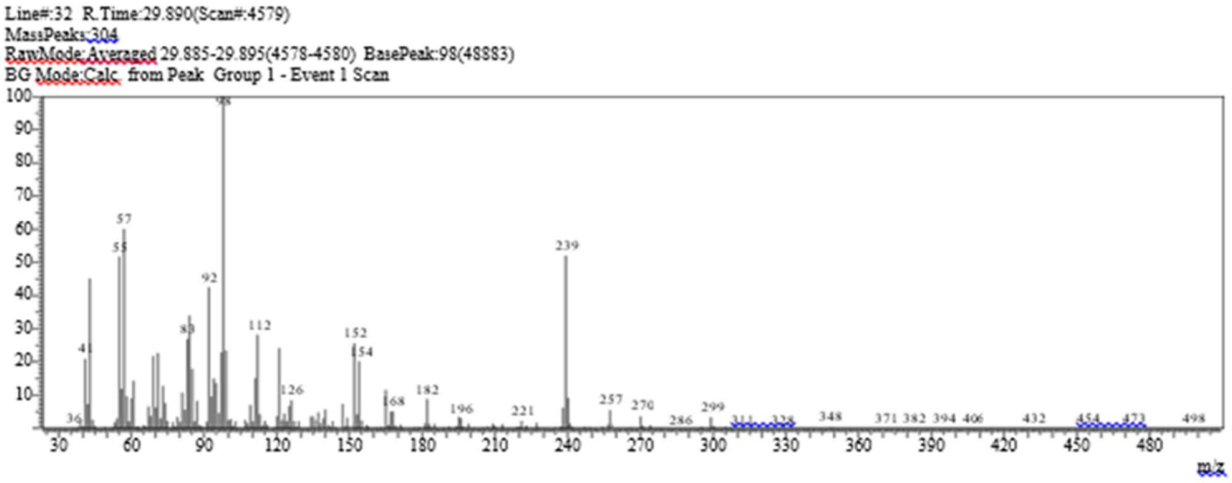
Graph 30: Chromatogram of Octadecanoic acid, ethyl ester



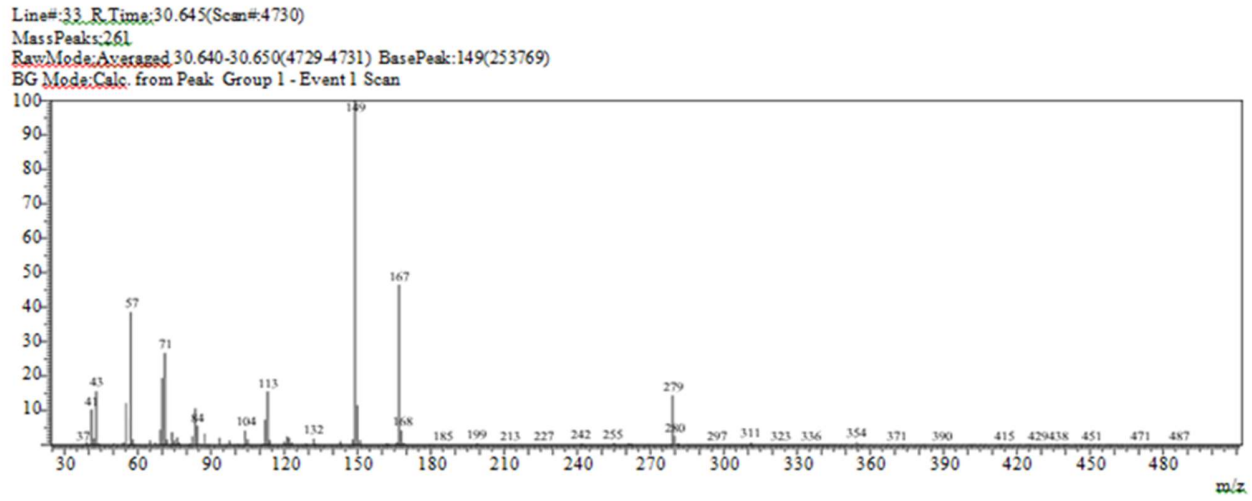
Graph 31: Chromatogram of Cyclohexane, tetradecyl



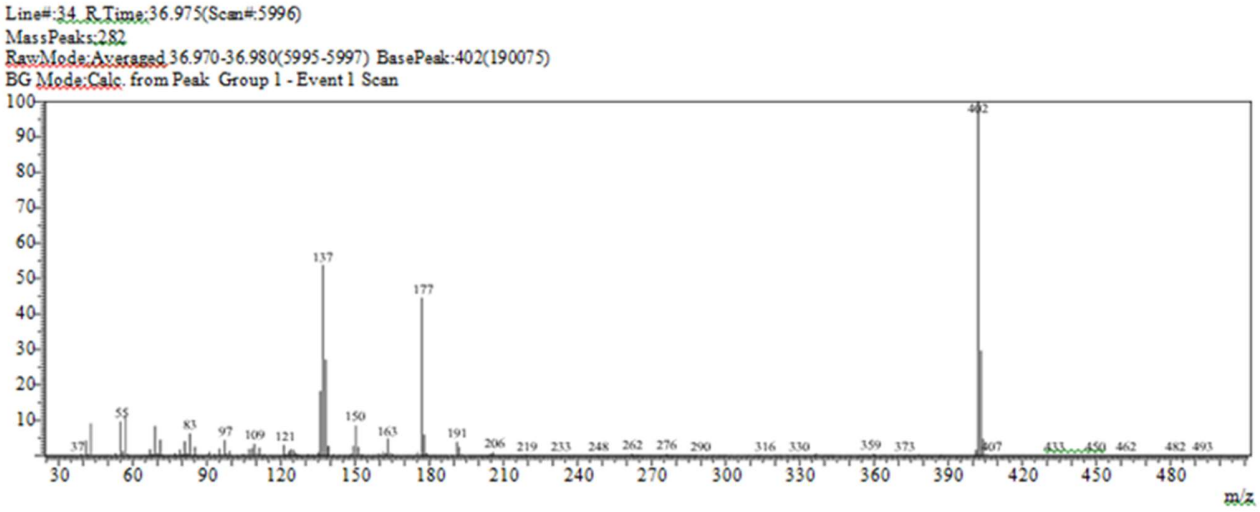
Graph 32: Chromatogram of 1-Hexacosanol



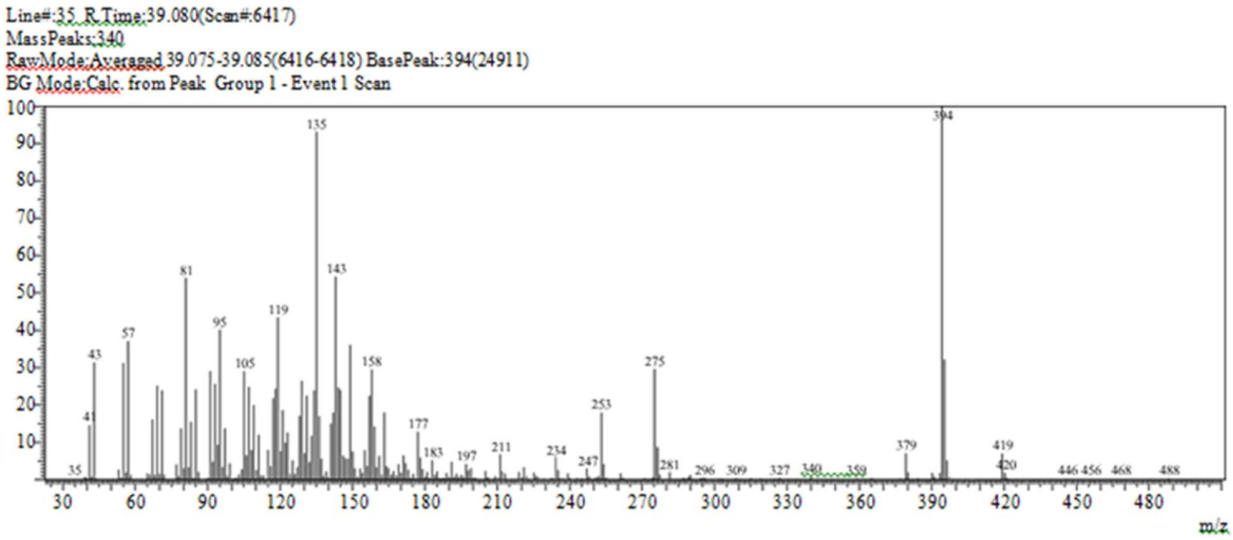
Graph 33: Chromatogram of Palmitoyl chloride



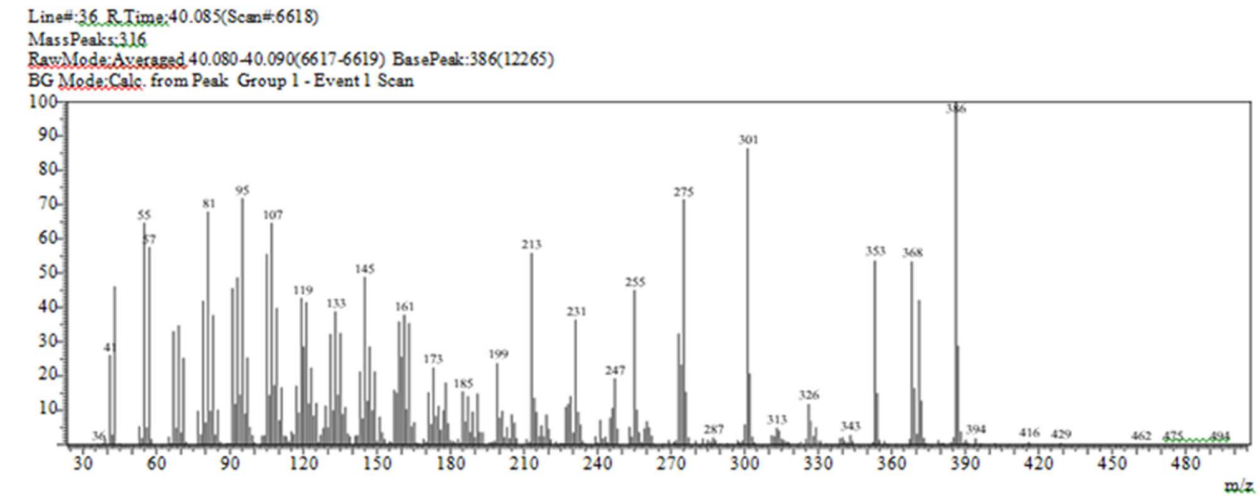
Graph 34: Chromatogram of Bis (2-ethylhexyl) phthalate



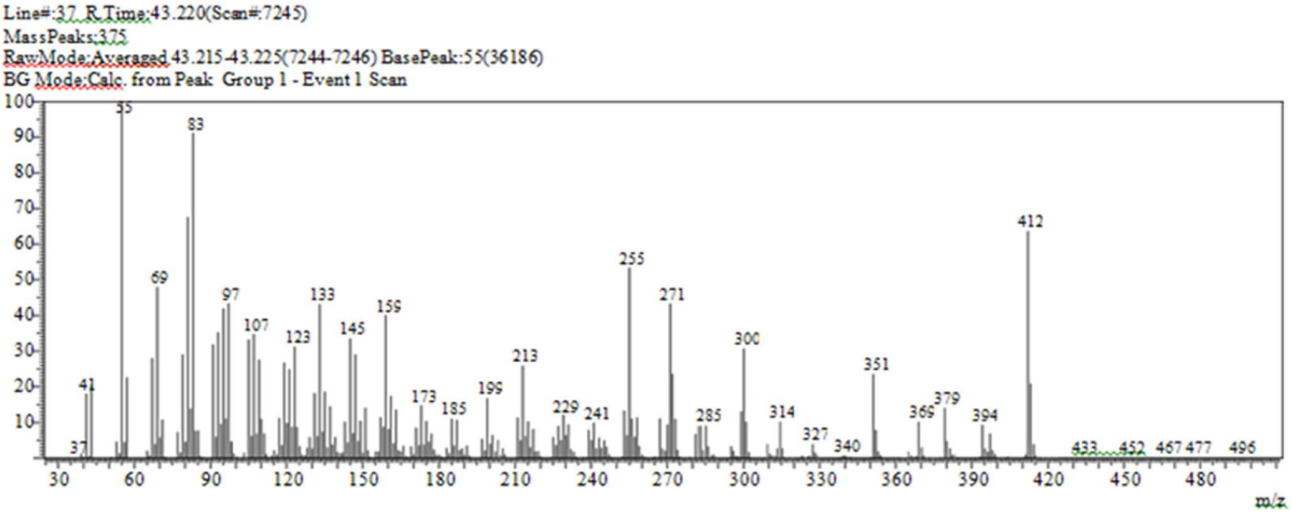
Graph 35: Chromatogram of delta. -Tocopherol



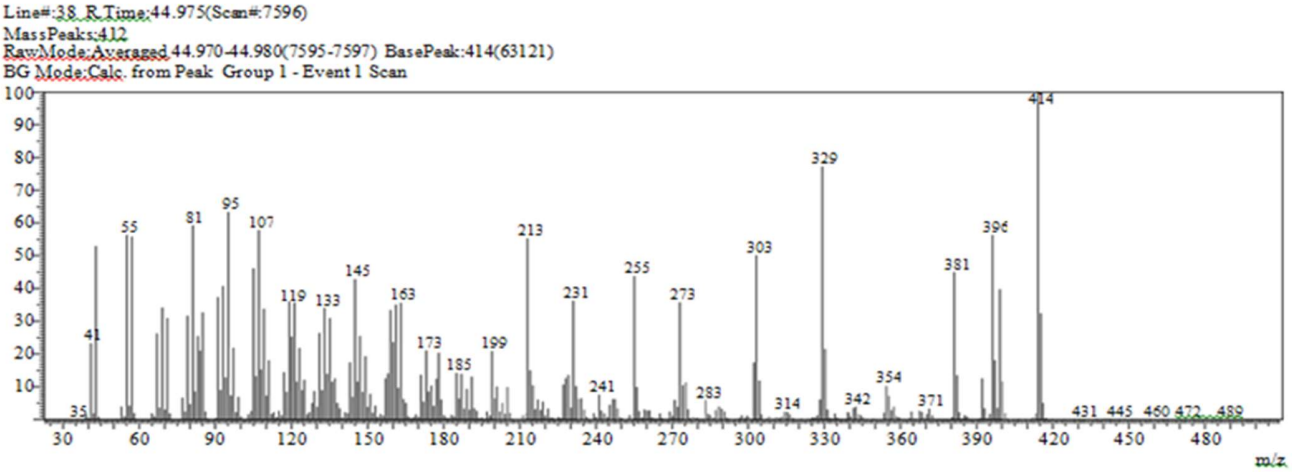
Graph 36: Chromatogram of Cholesta-4, 6-dien-3-ol, (3. beta.)-



Graph 37: Chromatogram of Cholesterol



Graph 38: Chromatogram of Stigmasterol



Graph 39: Chromatogram of gamma. -Sitosterol

The GC–MS analysis revealed a diverse phytochemical profile consisting of fatty acids, fatty acid esters, flavonoid, phenolic compounds, alcohols, sterols, and other bioactive metabolites. Major constituents identified include n-hexadecanoic acid (27.81%), DL-Proline, 5-oxo-, ethyl ester (10.77%), γ-sitosterol (7.94%), 9,12,15-octadecatrienoic acid (4.60%), and phytol (4.22%), which are known for their antimicrobial, antioxidant, anti-inflammatory, and potential therapeutic activities. The high proportion of saturated and unsaturated fatty acids, along with phytosterols and tocopherols, indicates significant nutritional and pharmacological potential. This compositional profile supports the traditional uses of the sample and suggests promising applications in food, cosmetic, and pharmaceutical formulations.

CONCLUSION

The GC–MS analysis of ethanolic extracts of *Allium fistulosum* leaves revealed a rich phytochemical profile comprising fatty acids, phenolic derivatives, phytosterols, tocopherols, and other biologically active compounds. The predominance of n-hexadecanoic acid, DL-Proline derivatives, and γ-sitosterol indicates strong pharmacological potential, particularly in antimicrobial, antioxidant, and anti-inflammatory applications. These results validate the traditional medicinal uses of *A. fistulosum* and emphasize its importance as a natural source of therapeutic agents. Further studies focusing on isolation, biological evaluation, and formulation development are recommended to fully exploit its medicinal and commercial potential.

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