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## Research Article



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## Phytochemical analysis of bio-active compounds in the Ethanolic seed extract of *Sesamum indicum* (L) by Gas Chromatography – Mass Spectrometry

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### Abstract

**Background:** Pharmacologically active constituents with physiological effects are known to be present in medicinal plants and its usage has been increased over the past decade. The aim of the present study was to identify the lead compounds present in the ethanolic seed extract of *Sesamum indicum* (L). **Methods:** Gas Chromatography – Mass Spectrometry (GC-MS) analysis of the ethanolic seed extract of *Sesamum indicum* (L). was carried out on a Perkin Elmer Turbo Mass Spectrophotometer (Norwalk, CT06859 and USA) which includes a Perkin Elmer Auto sampler XLGC. **Results:** The phytochemical test of the ethanolic seed extract of *Sesamum indicum* (L). shows the presence of alkaloids, steroids, flavonoids, saponins, phenols, tannins, amino acids, proteins and reducing sugars. The components present in the GCMS are palmitic acid, linolenic acid, methyl esterate, stearic acid, glycerin, and oleic acid. **Conclusion:** The synergistic activities are due to the phytochemicals rich nature of *Sesamum indicum* seeds.

**Keywords:** *Sesamum indicum*, seed, ethanolic extract, phytonutrients, GC-MS.

### Introduction

Natural goods are a vital source of therapeutic substances. Natural goods have been an essential component of history and culture for eons, serving as the foundation of traditional healing systems all across the world. Herbal medicine usage has been increased over the past decade and

are known to contain pharmacologically active constituents with physiological effects. In this regard, the seeds of *Sesamum indicum* is used traditionally for various ailments. The aim of the present study was to identify the lead compounds presents in ethanolic seed extracts of *Sesamum indicum* (EESI).

SESAME '*Sesamum indicum* L' is a commonly growing plant species mainly in tropical and subtropical regions in the world. Major producers of sesame are Burma, India, China Tanzania, Myanmar, Sudan, Ethiopia and Nigeria. Sesame seeds are used to prefferd in breads, biscuits and crackers, and as seasoning in food around the world (Namiki, 2007). This seed is a valuable oil seed crop and it is one of the oldest domesticated oilseed crops.

While the usual skin tones are black and white, the hue of sesame seeds ranged from cream-white to charcoal black. According to ancient Asian beliefs, black sesame seeds are healthier than white ones. This is why they are included in the People's Republic of China Pharmacopoeia (PPRC, 2015) as a benefit to the kidneys and liver in traditional Chinese medicine (TCM). It has been reported that the seed colors of sesame affect the phytochemical contents and their biological Activities (Zhang et al., 2013). Phytochemical compounds in sesame seed and antiproliferative activities of various sesame seeds will benefit the sesame seed planters, relevant manufacturers, and ordinary consumers such as a sesamol, anthrasesamol F and sesamol these are have been proved to have in vitro/in vivo antioxidant and antiaging activity (Zuo, et al., 2013; M. Tada, et al., 2013; Furumoto and Nishimoto, 2016). Moreover sesamol and sesamin are showed anti-inflammatory, antihypertensive, and anticarcinogenic effects in numerous studies (Hsu et al 2013; Xu et al., 2015). Sesame cultivars and landraces exhibit remarkable diversity, which is thought to be the consequence of farmers choosing different kinds and perhaps even of repeated domestication. In spite of its age and economical importance for local economies, in this is regarded as an orphan crop and research devoted to sesame has been scarce, for instance, sesame is not mandated by any international crop research center (Islam et al., 2016).

Oil of *Sesamum indicum* is valued for its sensory characteristics and resistance to rancidity. Sesame oil also exerts antioxidative activity and possesses in health promoting properties, which are attributed to tocopherols, tocotrienols and lignans

(Pathak et al., 2014). Bioactive compounds extracted from byproducts (agro-food chain) could be one of the promising ways to produce natural antioxidants (Azabou et al., 2020), including phenolics, flavonoids, carotenoids, saponins, and alkaloids (Silva et al., 2019). Major ligans of sesame or sesamin and sesamol. Total contents of these two ligans in sesame seed may exceed 1.4%

## Materials and Methods

### Collection and authentication of sample

The sesame seed was collected locally from Velachery, Chennai, Tamilnadu, INDIA. The collected sesame seed was authenticated (Reg num –EESI [(PARC/2021/4617) by Prof.P.Jayaraman, Ph.D. Director Institute of Herbal Botany, Chennai.

### Sample preparation

10g of sesame seed powder was dissolved in 100ml of 95% ethanol. Then the ethanolic extract was vacuum evaporated and stored for further analysis.

### Qualitative analysis of phytochemicals

#### Test for Proteins:

#### Ninhydrin Test:

To 1ml of extract drops of 5% ninhydrin solution was added heated in boiling water bath for 5 minutes Purple or bluish colour is observed.

#### Test for Tryptophan:

To 2ml of extract, few drops of glyoxalic acid and concentrated sulphuric acid were added along the sides of the test tubes. Reddish violet ring was formed in junction of two layers.

**Test for steroids:** To the extract 5ml of chloroform and equal volume of concentrated sulphuric acid and acetic acid was added by the sides of the test tubes. The upper layer turns red

and sulphuric acid layer turned yellow with green fluorescence.

### Test for Alkaloids:

#### Wagner's Test:

To 2 ml of extract few drops of wagner's reagent was added along the sides of the test tubes. A reddish brown precipitate indicates the presence of alkaloids.

#### GC-MS Analysis

A Perkin Elmer Turbo Mass Spectrophotometer (Norwalk, CTO6859, and USA) with a Perkin Elmer Auto sampler XLGC was used for the GC-MS analysis. The column used was Perkin Elmer Elite - 5 capillary column measuring 30m × 0.25mm with a film thickness of 0.25mm composed of 95% Dimethyl polysiloxane. At 0.5 milliliters per minute, helium was utilized as the carrier gas. 1µl sample injection volume was utilized. The inlet temperature was maintained as 250°C. First, the oven temperature was set to 110°C for four minutes, and then it was raised to 240°C. then set to rise to 280°C at a pace of 20°C per minute for the final five minutes. Total run time was 90 min. A temperature of 200°C was maintained in the MS transfer line. The source temperature was maintained at 180°C. Using electron impact ionization at 70 eV, GCMS data was processed, and total ion count (TIC) was used to assess the data for compound identification and quantification. The component spectrums were compared to the database of known component spectra kept in the GC-MS library. Data processing and peak area measurement were done using Turbo-Mass OCPTVS-Demo SPL software.

## Results and Discussion

Ayurvedic system of medicine is the natural boon to human population which provides natural and harmless protection against major ailments. The phytonutrients formed as secondary metabolites of plant products consumed in the diet, play an important role in maintaining good health.

## Phytochemical analysis

The phytochemical analysis was carried out in *Sesamum indicum* seed extracts obtained using ethanol. As shown in table 1 ethanolic seed extract of *Sesamum indicum* shows the presence of alkaloids, steroids, flavonoids, saponins, phenols, tannins, amino acids, proteins and reducing sugars. Phytochemicals such as saponins, terpenoids, flavonoids and alkaloids have anti-inflammatory activities (Bhaskar and Balakrishnan, 2009). *Sesamum indicum* has high amount of tannins and they play an important role in the treatment of intestinal disorders like diarrhoea.

Flavonoids are phenolic chemicals that have been extracted from more than 8000 different types of vascular plants. They act in plants as antioxidants, antimicrobials, photoreceptors etc. Flavonoids exhibits biological activities, including antiallergenic, antiviral, anti-inflammatory and vasodilating actions (Marsh and Steinbeck, 2010). They are also beneficial to the plant itself as stress preventing agents as attractants as feeding deterrents and in general by their significant role in plant resistance against infection.

Phytochemicals classified as phenolic compounds comprise the largest group and are primarily responsible for plants' antioxidant action. Many naturally occurring triterpenoids exhibited a good anti-inflammatory activity and they have been isolated from various plants. There is a growing interest in natural triterpenoids due to their bactericidal, fungicidal, antiviral, cytotoxic and anticancer properties.

Phenol is a type of organic compound. Plant based compounds containing phenol are known to be antioxidant. They are strong antioxidant and might prevent oxidative damage to biomolecules such as DNA, lipids and proteins which play a role in chronic diseases such as cancer and cardiovascular disease. It can stop the reaction of free radicals with other molecules in your body.

Alkaloids, flavonoids, phenols and tannins are responsible for the antioxidant property of the

plant. The presence of these phytochemicals supports the medicinal use of *Sesamum indicum*.

**Table 1: Qualitative analysis of phytochemical in *Sesamum indicum* extracts**

S.No	Phytochemicals	Methanol	Ethanol	Petroleum Ether
1.	Test of Alkaloids	+	+	+
2.	Test of Saponins	+	+	-
3.	Test of Phenols	+	+	-
4.	Test of Flavonoids	+	+	-
5.	Test of Ferric Chlorides	+	+	-
6.	<b>Amino acids</b> Ninhydrin	+	+	+
7.	Tryptophan	+	+	+

#### GC-MS Analysis of ethanolic seed extract of *Sesamum indicum* (L.)

The seed kernels of *Sesamum indicum* L. (Family: Pedaliaceae) were extracted with ethanol and yield of components determined by Gas Chromatography/Mass Spectrometry (GC/MS). The free radical scavenging activities of ethanolic extract against 1, 1-Diphenyl-2-picrylhydrazyl (DPPH) were determined by UV spectrophotometer at 517 nm. Phytochemical screening revealed the presence of numerous bioactive compounds including steroids, phenolic, terpenoids, fatty acids and different types of ester compounds. The ethanolic extract was purified and analyzed by GC MS. The prevailing compounds found in ethanolic extract were Carvacrol (0.04%), Sesamol (0.11%), 4-Allyl-2-methoxy-phenol (0.04%), Palmitic acid (1.08%),

cis-9-Hexadecenal (85.40%), Lineoleoyl chloride (0.52%), Palmitic acid -monoglyceride (0.40%), Dihydro-aplotaxene (0.61%), Oleoyl chloride (1.11%), (+)-Sesamin (4.73%), 1,3-Benzodioxole, 5-[4-(1,3-benzodioxol-5-yloxy)tetrahydro-1 H,3 H-furo [3,4-c]furan-1-yl], [1 S-(1,3,4,6 .), (2.01%)], 6-Nitrocholest-5-en-3-yl acetate (0.22%), Ergost-5-en-3 -ol (2.35%) and 24-Propylidenecholesterol (0.16%). The presence of saturated and unsaturated fatty acids in ethanolic extract justifies the use of this plant to treat many ailments in folk and traditional medicine. Ethanolic extract have shown significant antioxidant activity ( $IC_{50} 120.38 \pm 2.8 \mu\text{g/ml}$ ). The presence of phenolic (Sesamol), lignin (Sesamin) compounds and unsaturated fatty acids are reported as possible contributor for antioxidant activity of seed extract.

EESI [Sesame seed] Ethanol extract.

Name: Dimethyl Sulfoxide [Formula: C<sub>2</sub>H<sub>6</sub>OS]

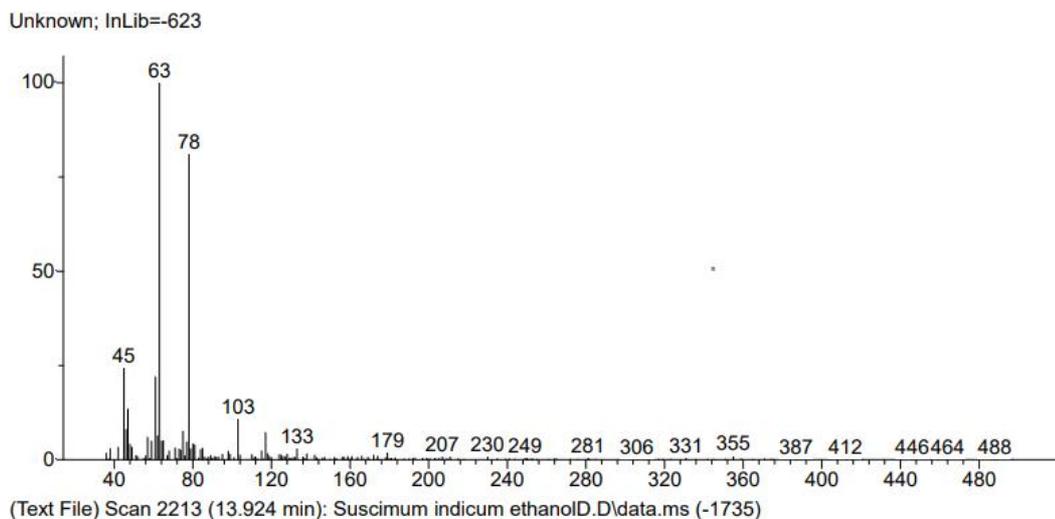


Fig 1.1 Ethanolic seed extract of *Sesamum indicum*

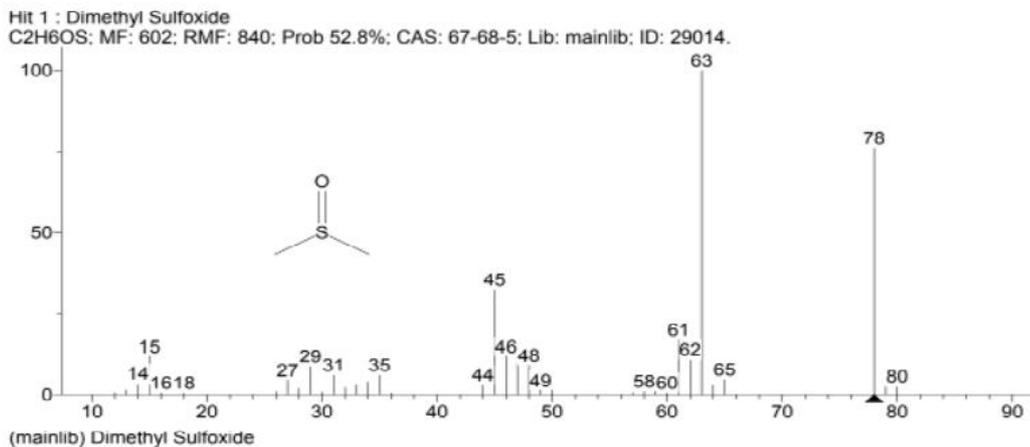


Fig 1.2 Standard

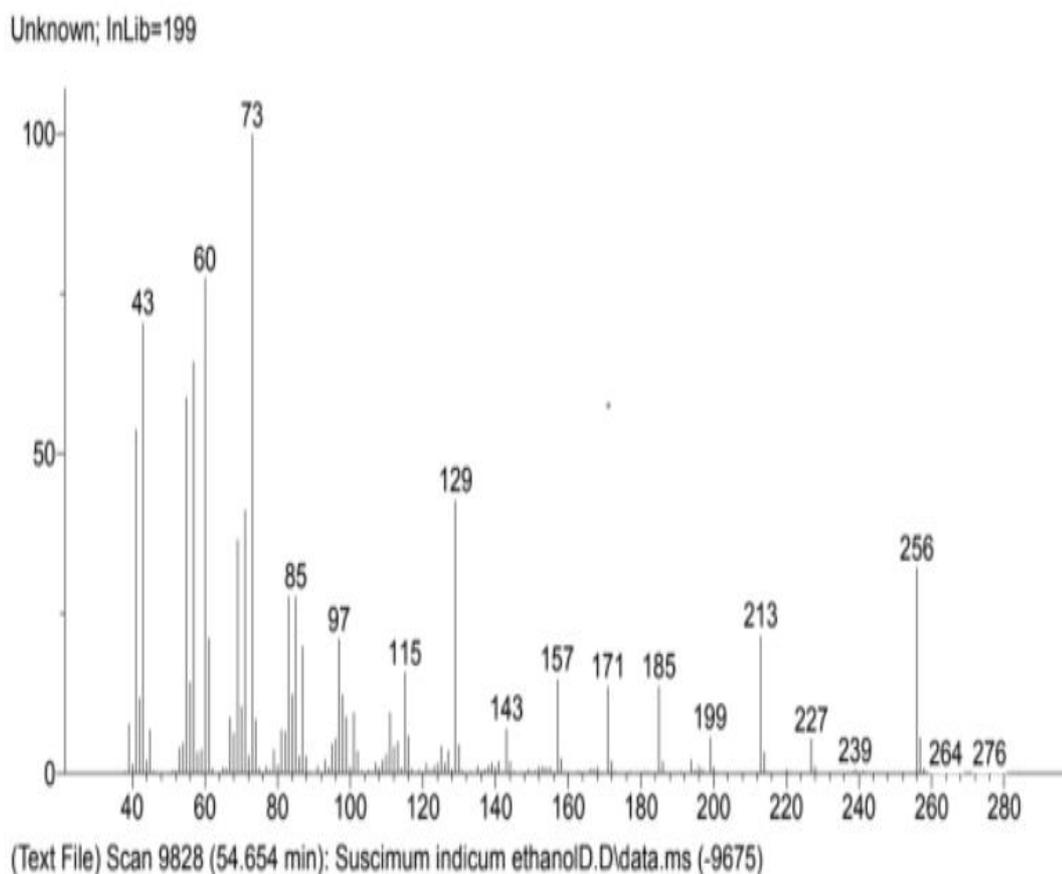
Dimethyl Sulfoxide is a highly polar organic liquid that is used widely as a chemical solvent and a free radical scavenger. It shows a range of pharmacological activity including analgesia and anti-inflammation.

Dimethyl sulfoxide (DMSO) is a chemical solvent that is sometimes used to help reduce inflammation and pain, and may also be beneficial in reducing leakage during chemotherapy treatment.

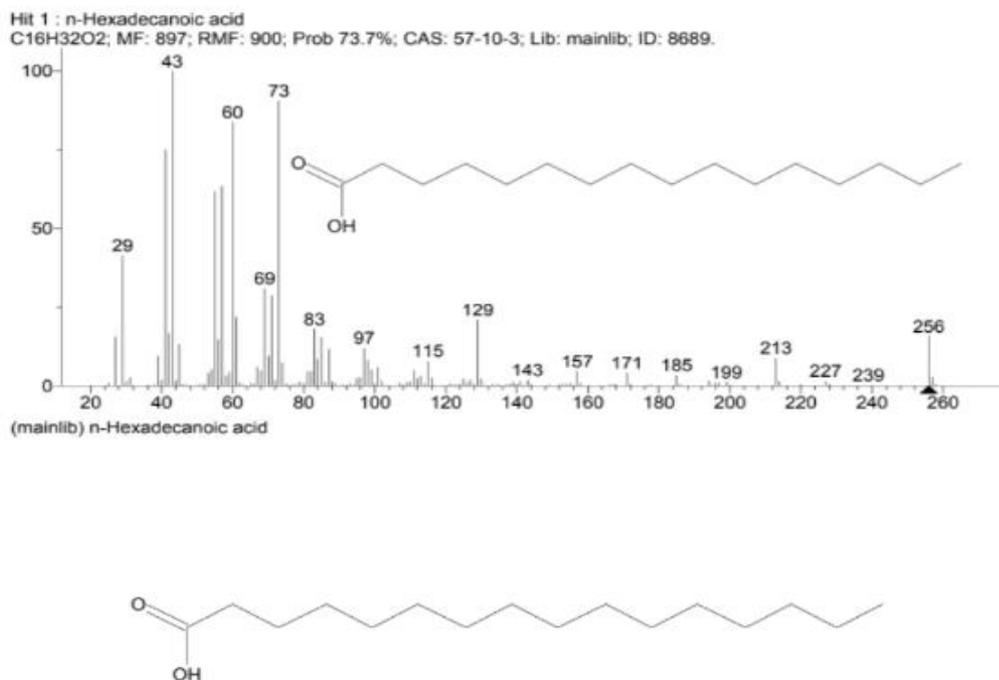
DMSO is used topically to decrease pain and speed the healing of wounds, burns, and muscle and skeletal injuries.

**Name: n-Hexadecanoic acid**

**Formula: C<sub>16</sub>H<sub>32</sub>O<sub>2</sub>**



**Fig 2.1** Ethanolic seed extract of *Sesamum indicum*



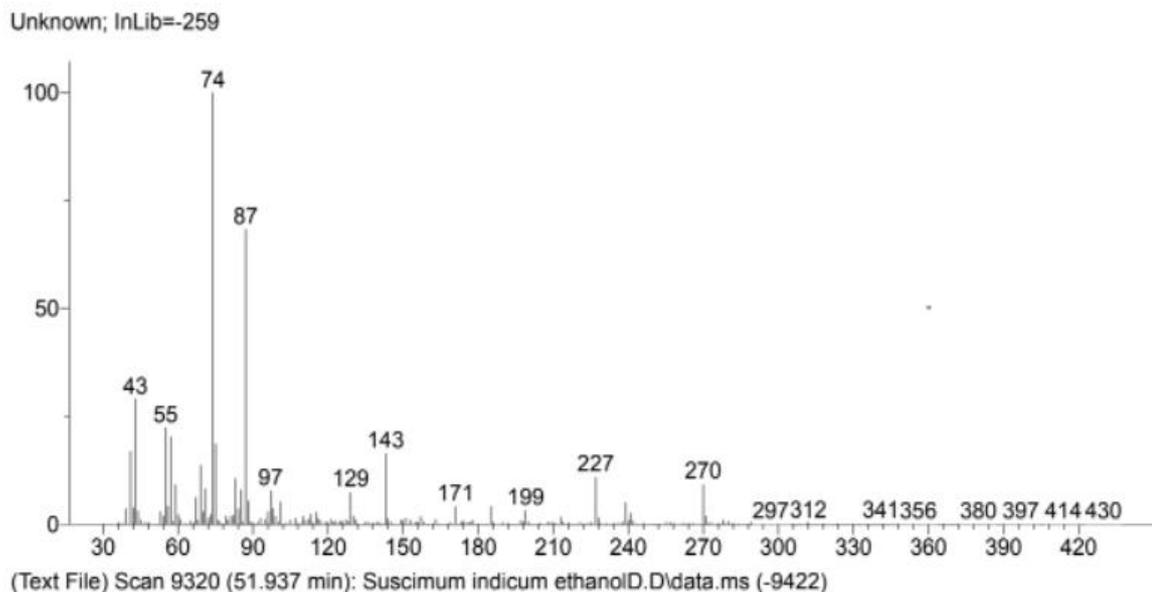
**Fig 2.2 Standard**

Palmitic acid, also called hexadecanoic acid, is one of the most common saturated fatty acids found in animals and plants.

The inferences from the present study validate the rigorous use of medicated oils rich in *n*-hexadecanoic acid for the treatment of rheumatic symptoms in the traditional medical system of India, Ayurveda

**Name: Hexadecanoic acid, ethyl ester**

**Formula: C<sub>18</sub>H<sub>36</sub>O<sub>2</sub>.**



**Fig 3.1 Ethanolic seed extract of *Sesamum indicum***

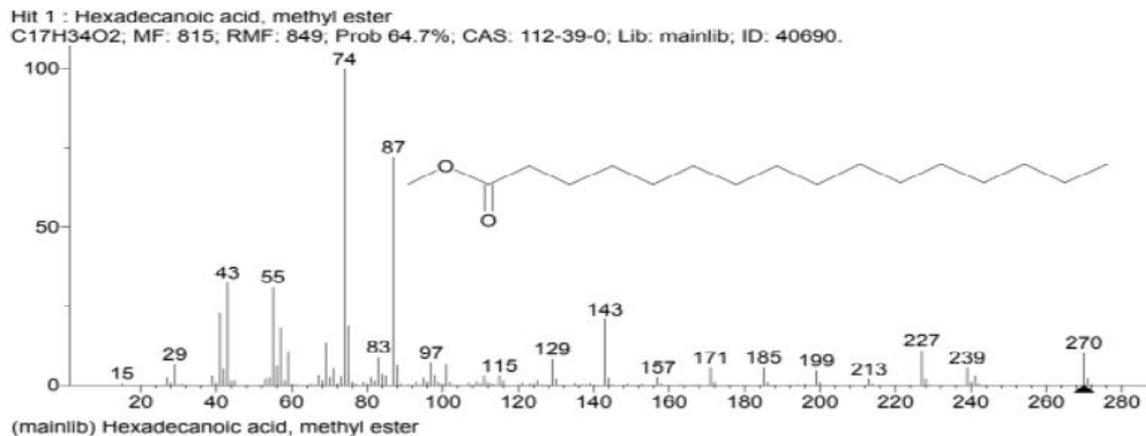


Fig 3.2 Standard

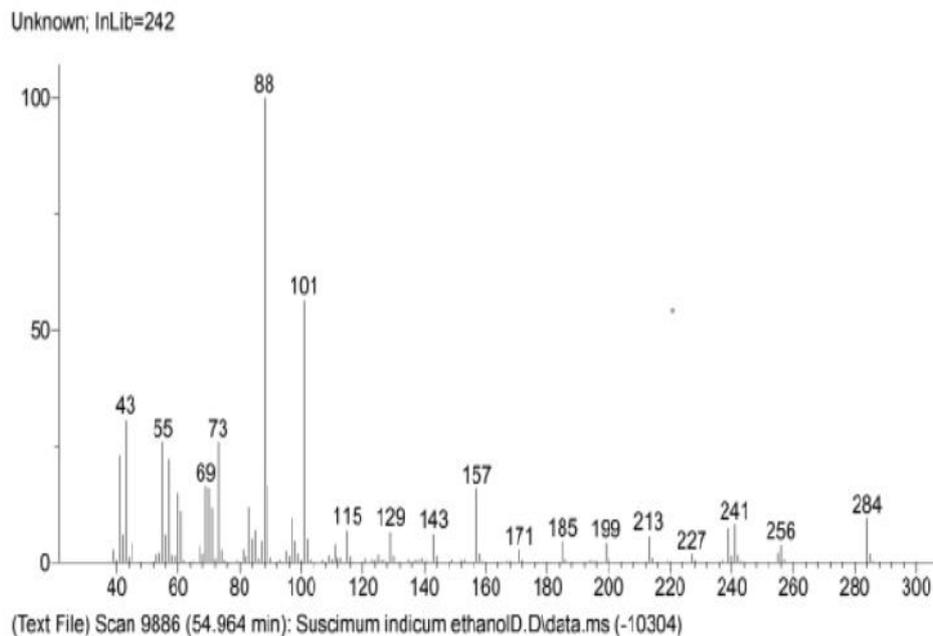
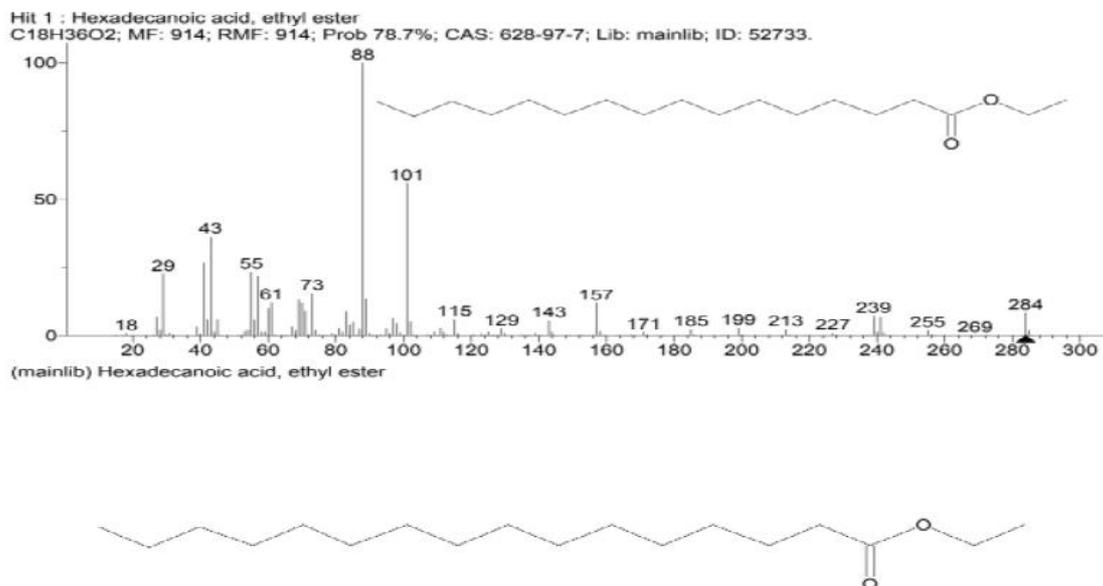


Fig 4.1 Ethanolic seed extract of *Sesamum indicum*

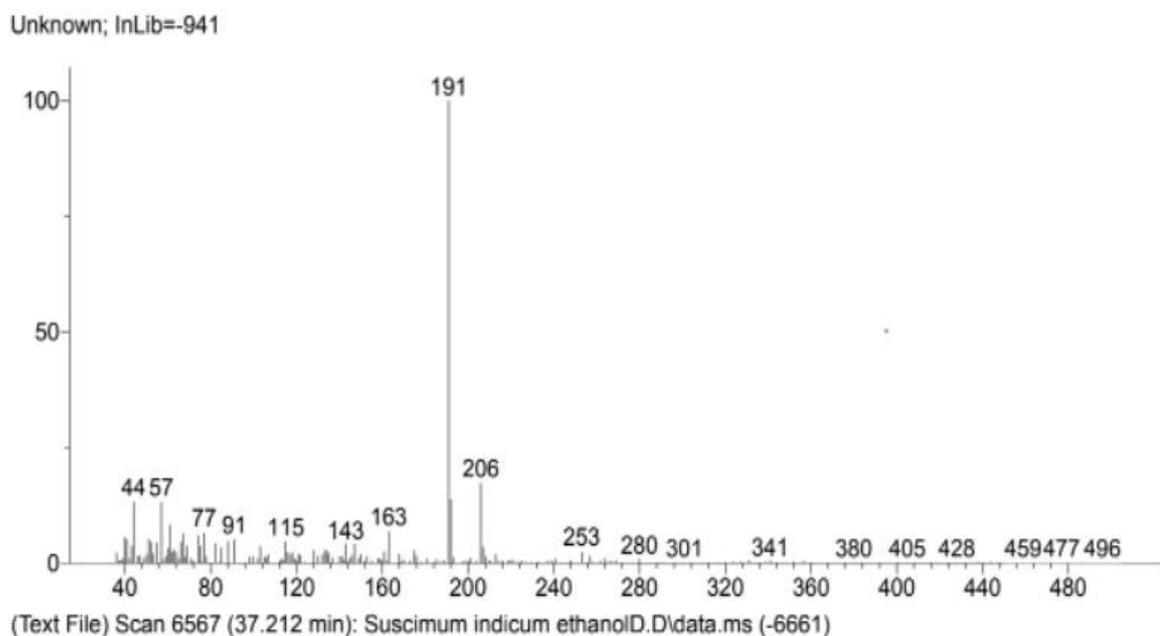


**Fig 4.2 Standard**

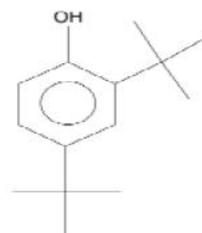
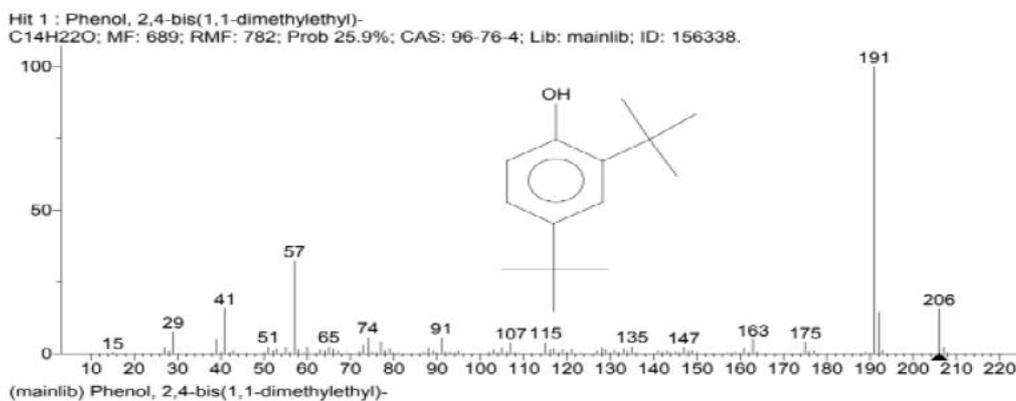
Ethyl hexadecanoate is a long-chain fatty acid ethyl ester resulting from the formal condensation of the carboxy group of palmitic acid with the hydroxy group of ethanol. It has a role as a plant metabolite. It is a hexadecanoate ester and a long-chain fatty acid ethyl ester.

Hexadecanoic acid, ethyl ester act as Antifungal, Antitumour, Anti-bacterial activity and anti-inflammatory property.

**Name: Phenol, 2,4-bis(1,1-dimethylethyl)-**  
**Formula: C<sub>14</sub>H<sub>22</sub>O.**



**Fig 5.1 Ethanolic seed extract of *Sesamum indicum***



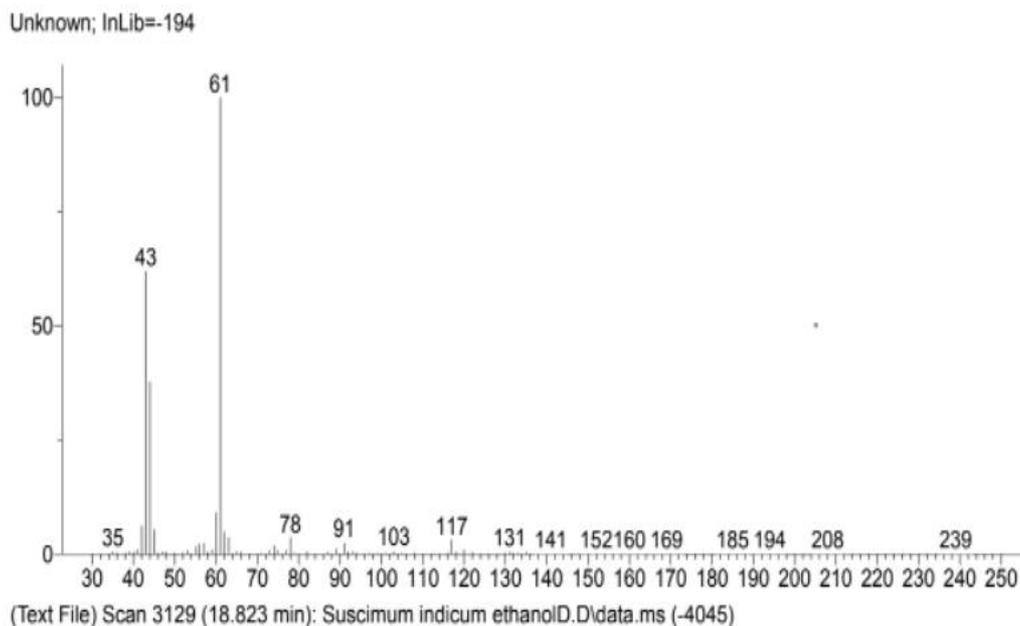
**Fig 5.2 Standard**

Phenolics are the most diverse group of secondary metabolites and mostly act as antioxidants, i.e., they are able to scavenge free radicals. Phenol-2,4-bis(1,1dimethylethyl) has been reported for anti-inflammatory activity as well as antioxidant

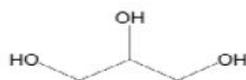
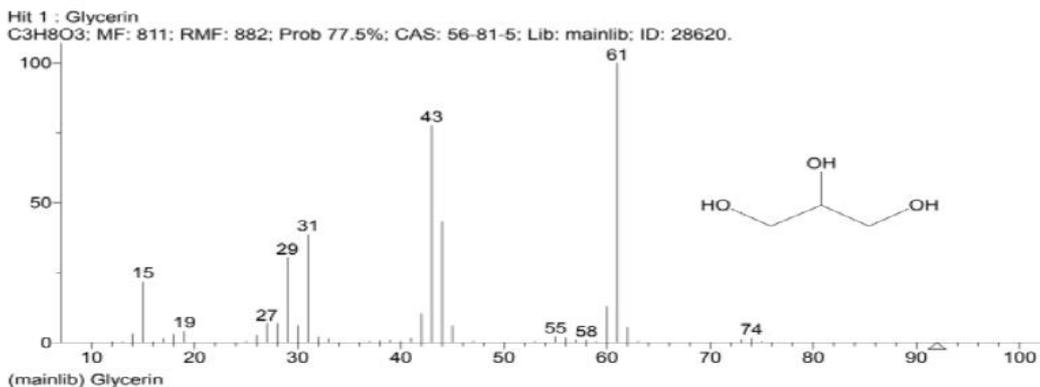
activity [24,39], and other identified phenolic compounds with antioxidant activity are phenol-2,6-dimethoxy and pyrogallol [41]. Pyrogallol has prominent hepatoprotective activity.

**Name: Glycerin**

**Formula: C<sub>3</sub>H<sub>8</sub>O<sub>3</sub>.**



**Fig 6.1 Ethanolic seed extract of *Sesamum indicum***



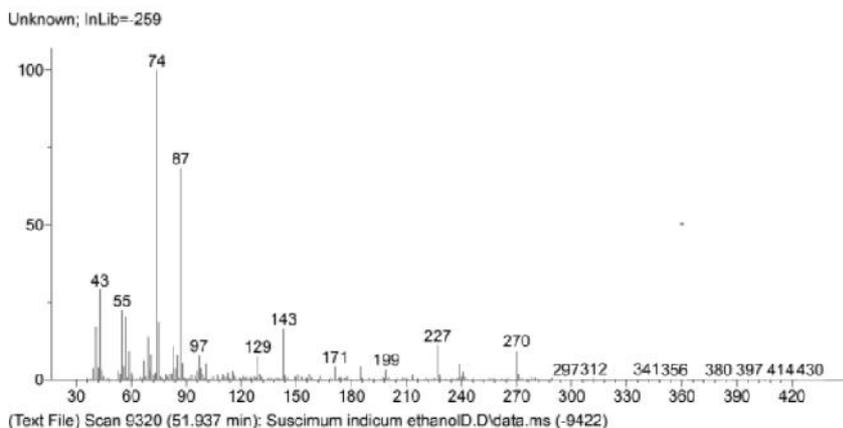
**Fig 6.2 Standard**

Glycerine is one of the most effective ingredients to treat dehydrated and dry skin. It can be found in almost any household. It is also known as glycerol and it can be extracted from vegetable fat as well as animal fat. This thick, odourless liquid

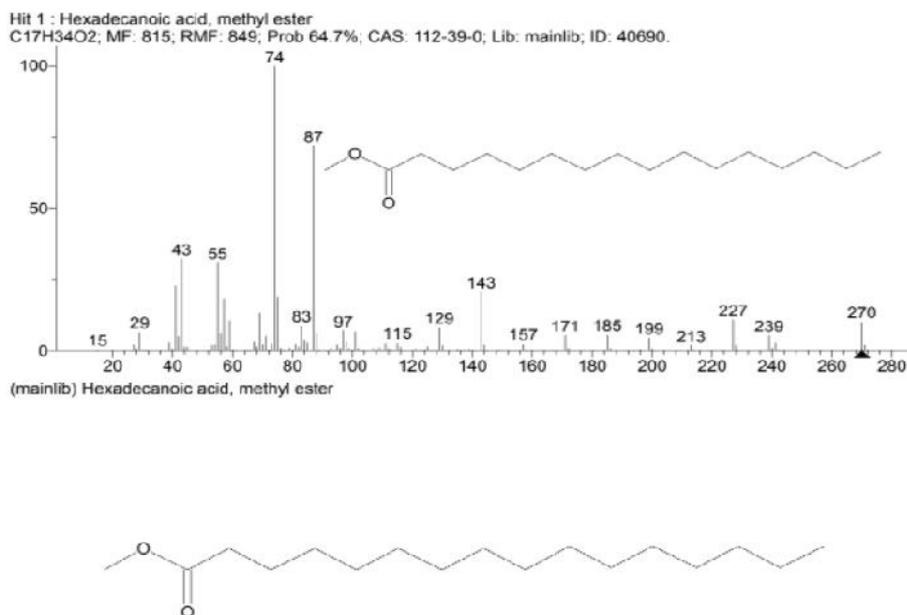
is used in various cosmetic products. Glycerol is a naturally occurring alcohol. It is an odorless liquid that is used as a solvent, sweetening agent, and also as medicine.

**Name: Hexadecanoic acid, methyl ester**

**Formula:  $C_{17}H_{34}O_2$**



**Fig 7.1 Ethanolic seed extract of *Sesamum indicum***



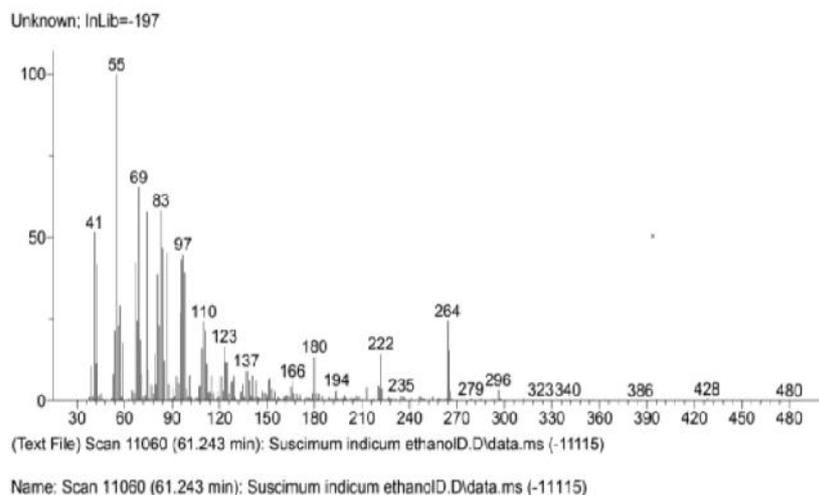
**Fig 7.2 Standard**

Methyl hexadecanoic acid, also known as palmitic acid methyl ester, belongs to the class of organic compounds known as fatty acid methyl esters. Methyl palmitate is a fatty acid methyl ester.

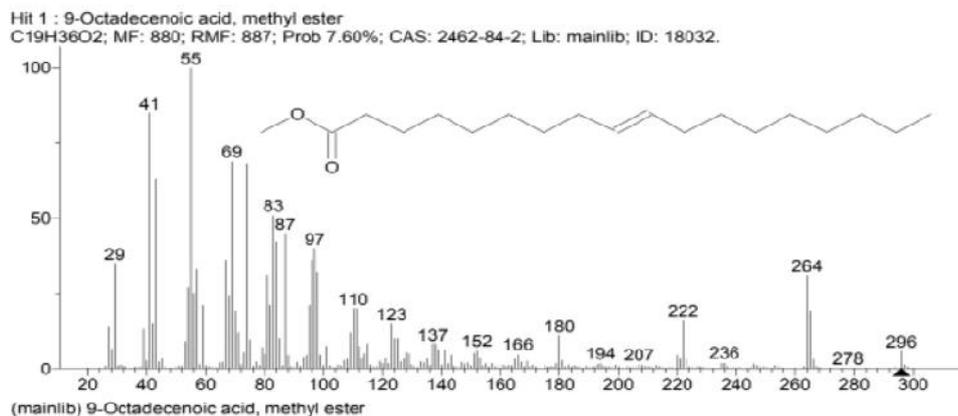
It has a role as a metabolite. Methyl esters of fatty acids produced from edible fats and oils are approved for some uses as direct food additives or as a supplementary source of fat.

The methyl esters of vegetable oils are excellent solvents for inks, polymers, and oils, with low volatility and good solubility.

**Name: 9-Octadecenoic acid, methyl ester.**  
**Formula: C<sub>19</sub>H<sub>36</sub>O<sub>2</sub>.**



**Fig 8.1 Ethanolic seed extract of *Sesamum indicum***



**Fig 8.2 Standard**

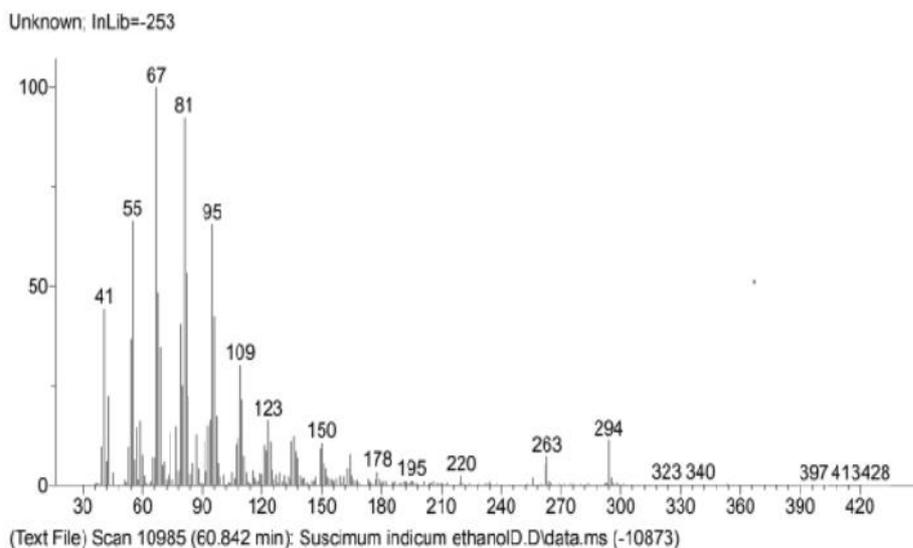
Stearic acid is a long-chain fatty acid. It is common in a variety of natural sources. Methyl oleate is a fatty acid methyl ester resulting from the formal condensation of the carboxy group of oleic acid with methanol. It has 31% mono unsaturated fatty acid in 9-Octadecenoic acid methyl.

### Oleic Acid

Oleic acid is classified as a monounsaturated omega-9 fatty acid, Preventing heart disease reducing cholesterol. It is also used for preventing cancer.

**Name: 9, 12-Octadecadienoic acid (Z, Z)-, methyl ester**

**Formula: C<sub>19</sub>H<sub>34</sub>O<sub>2</sub>.**



**Fig 9.1 Ethanolic seed extract of *Sesamum indicum***

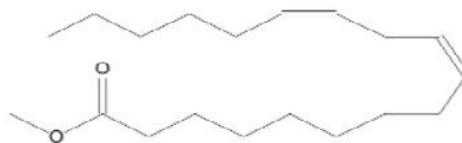
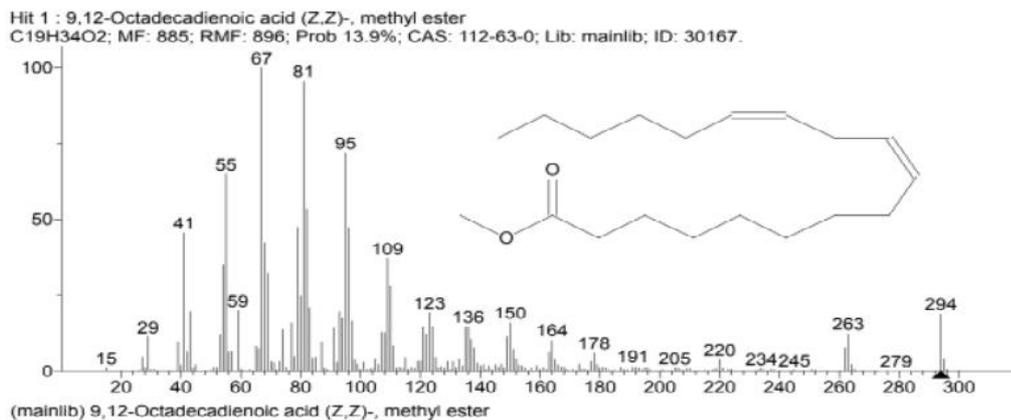


Fig 9.2 Standard

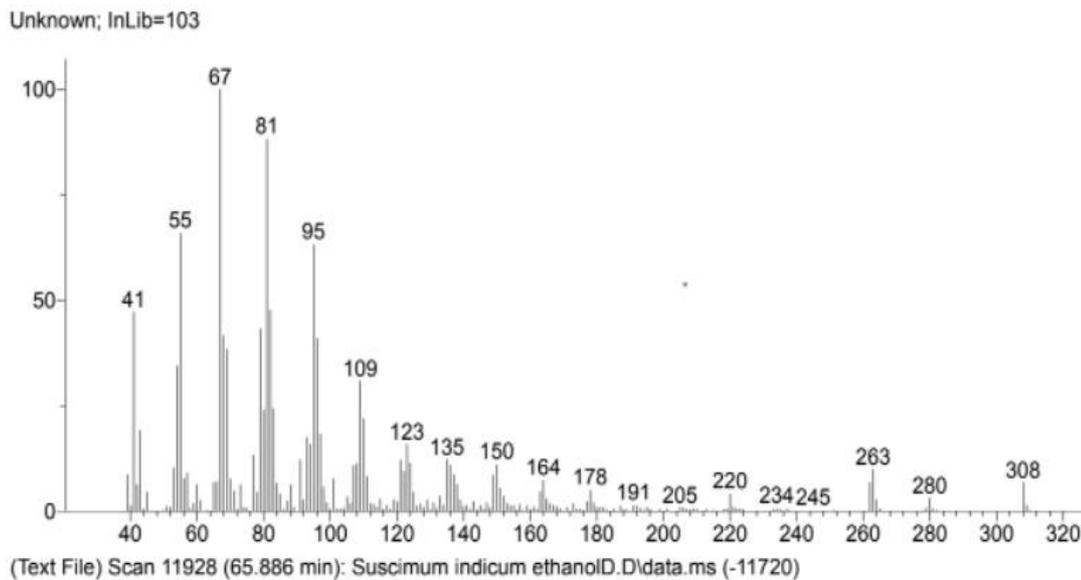
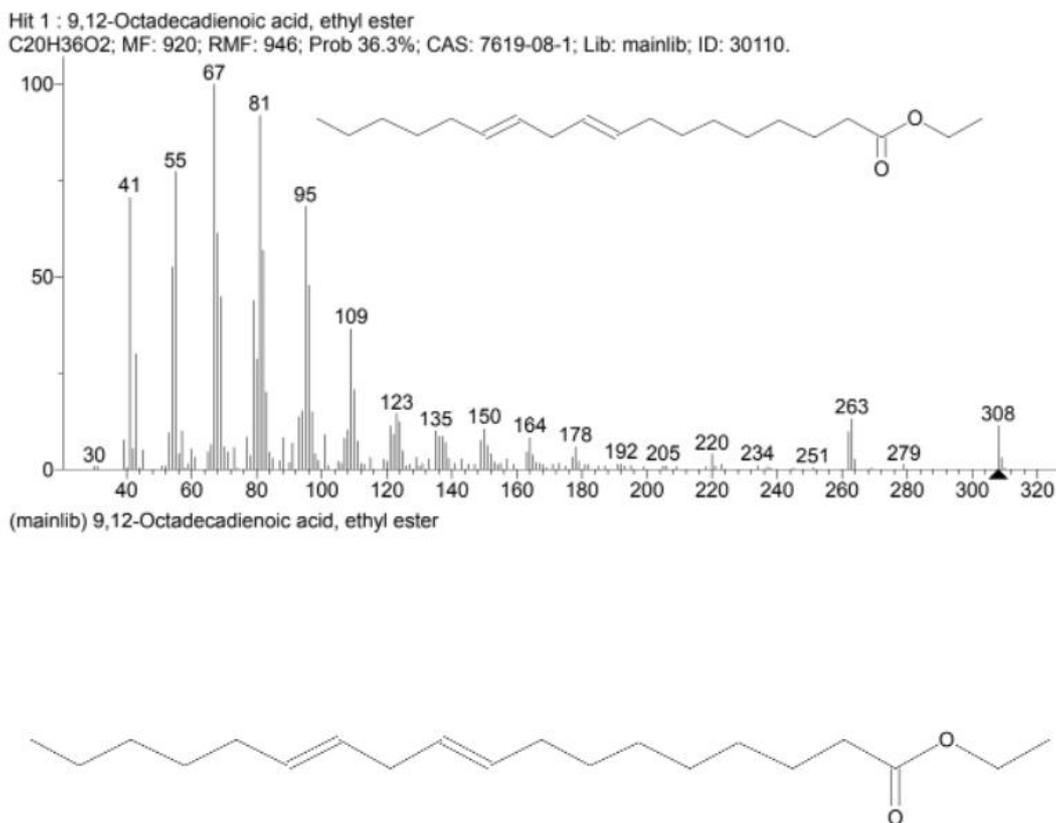


Fig 10.1 Ethanolic seed extract of *Sesamum indicum*



**Fig 10.2 Standard**

**Linoleic acid.**

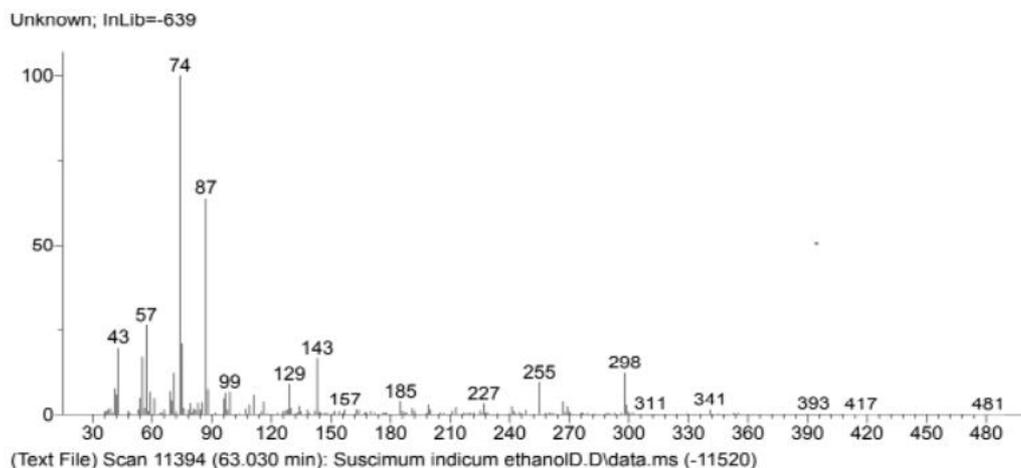
Linolenic Acid is an essential fatty acid belonging to the omega-3 fatty acids group. It is highly concentrated in certain plant oils and has been reported to inhibit the synthesis of prostaglandin resulting in reduced inflammation and prevention of certain chronic diseases.

Linoleic Acid is a polyunsaturated essential fatty acid found mostly in plant oils.

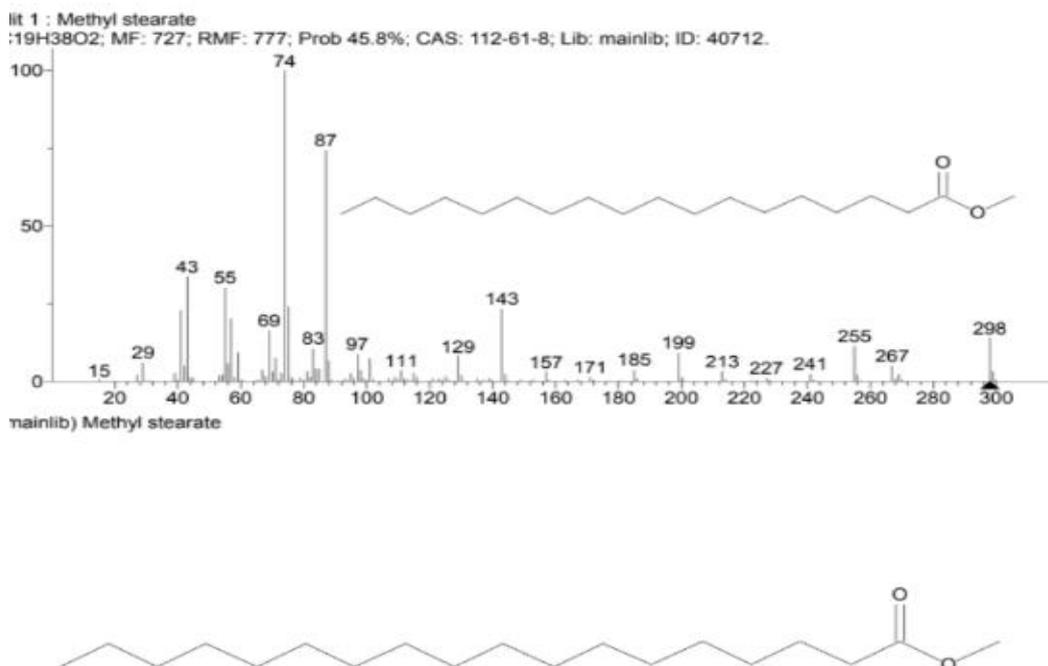
Alpha-linolenic acid is thought to decrease the risk of heart disease by helping to maintain normal heart rhythm and pumping. It might also reduce blood clots.

**Name: Methyl stearate**

**Formula: C<sub>19</sub>H<sub>38</sub>O<sub>2</sub>.**



**Fig 11.1 Ethanolic seed extract of *Sesamum indicum***



**Fig 11.2 Standard**

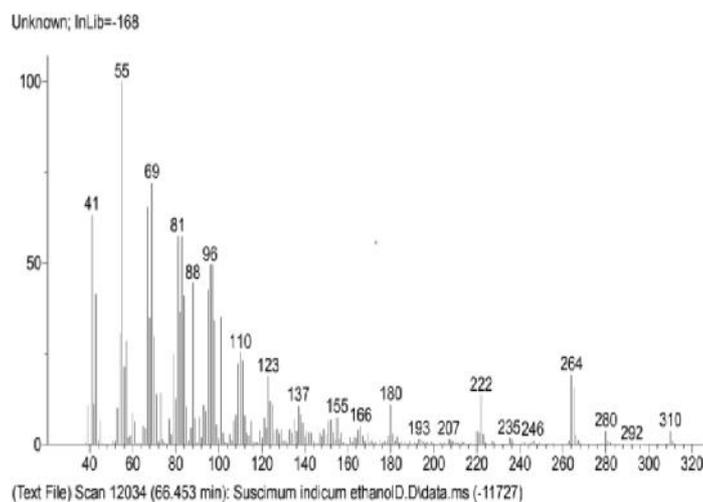
Methyl stearate is a fatty acid methyl ester and an octadecanoate ester. It has a role as a metabolite.

Methyl stearate is one of the key fatty acid methyl ester component of biodiesel fuel produced by the transesterification of triglyceride oil and a primary alcohol.

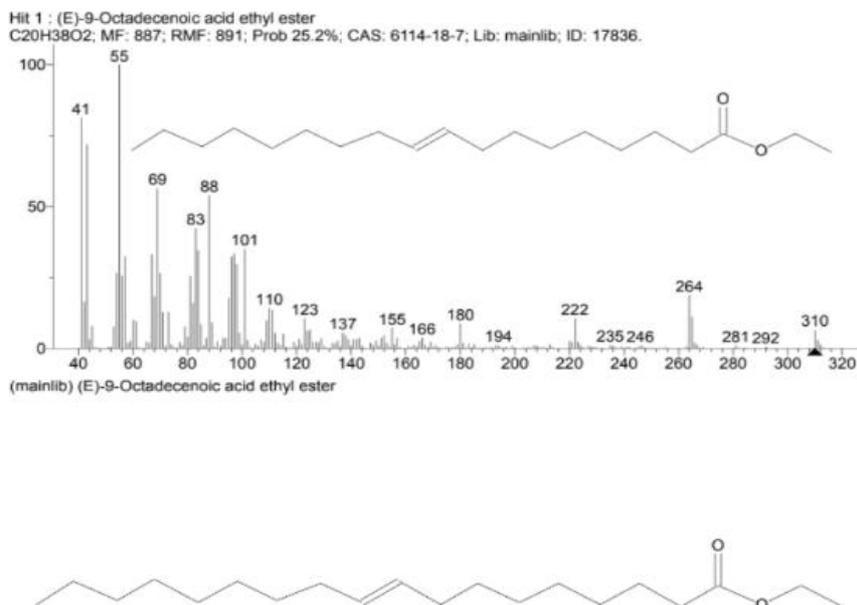
The deoxygenation of methyl octanoate and of methyl stearate on alumina-supported platinum was investigated with vapor-phase and with liquid-phase reactants in a flow reactor and in the semibatch reactor, respectively.

**Name: (E)-9-Octadecenoic acid ethyl ester**

**Formula: C<sub>20</sub>H<sub>38</sub>O<sub>2</sub>**



**Fig 12.1 Ethanolic seed extract of *Sesamum indicum***



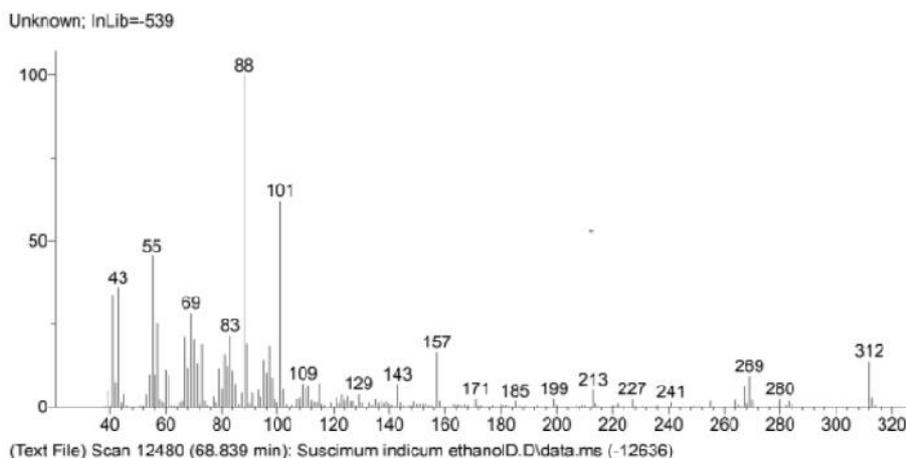
**Fig 12.2 Standard**

Ethyl oleate is a long-chain fatty acid ethyl ester resulting from the formal condensation of the carboxy group of Oleic Acid with

the hydroxy group of ethanol. It has a role as a plant metabolite and an acaricide. It derives from an Oleic Acid.

**Name: Octadecanoic acid, ethyl ester**

**Formula: C<sub>20</sub>H<sub>40</sub>O<sub>2</sub>.**



**Fig 13.1 Ethanolic seed extract of *Sesamum indicum***

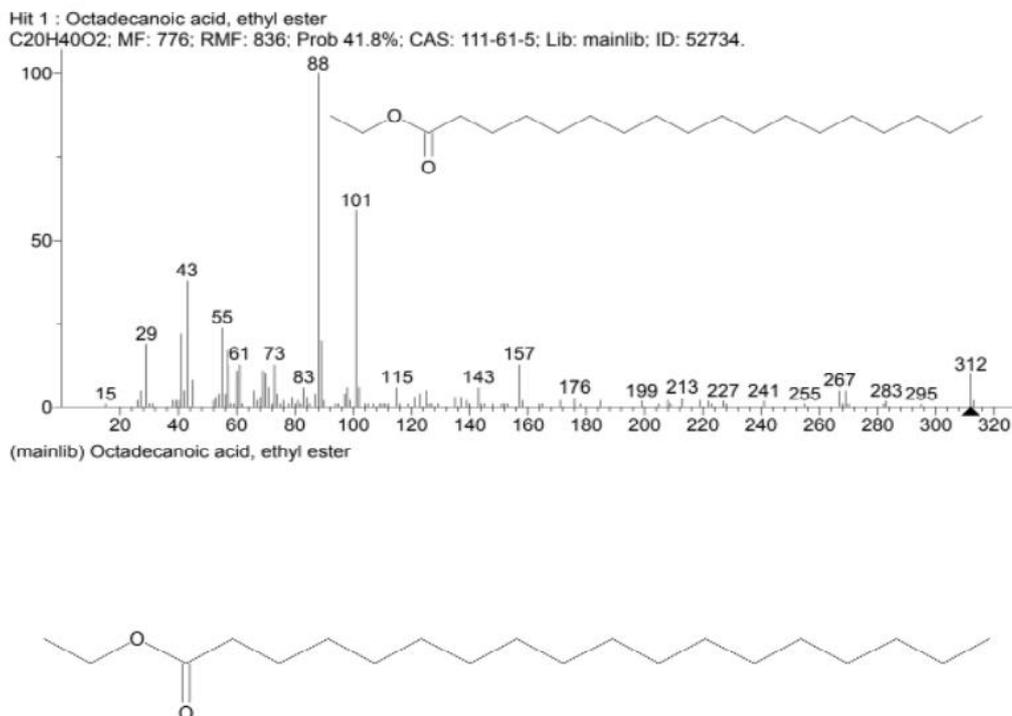


Fig 13.2 Standard

Ethyl octadecanoate is an octadecanoate ester obtained by formal condensation between the carboxy group of octadecanoic (stearic) acid and

the hydroxy group of ethanol. It has a role as a plant metabolite. It is a long-chain fatty acid ethyl ester and an octadecanoate ester.

**Name: 9,12-Octadecadienoic acid (Z,Z)-**

**Formula: C<sub>18</sub>H<sub>32</sub>O<sub>2</sub>**

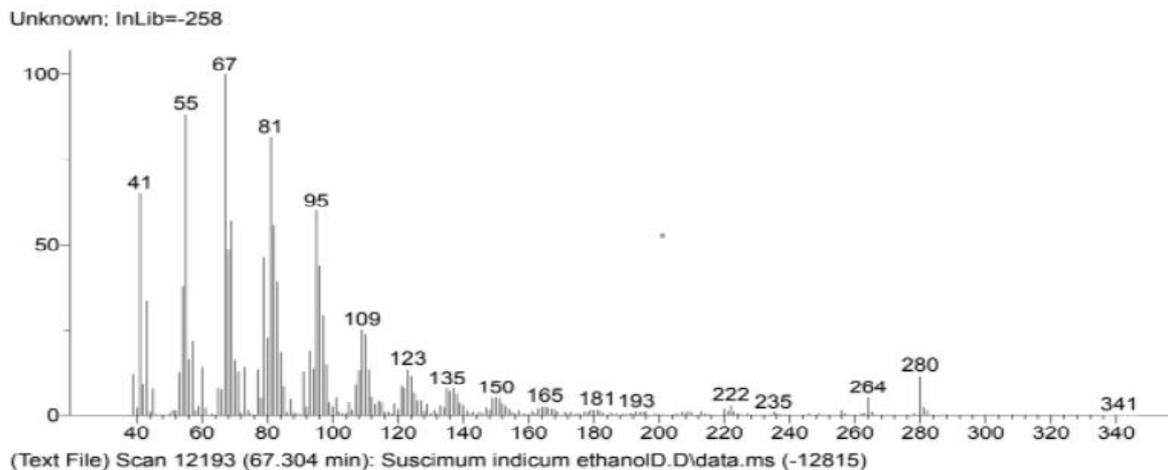


Fig 14.1 Ethanolic seed extract of *Sesamum indicum*

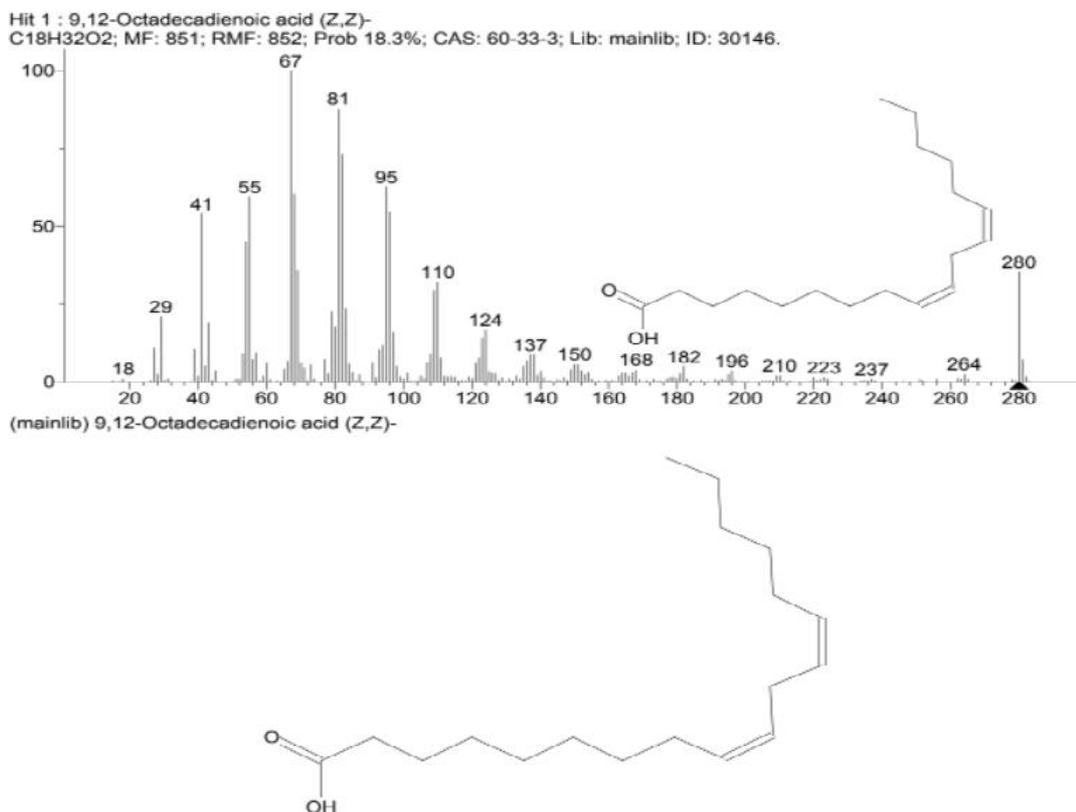


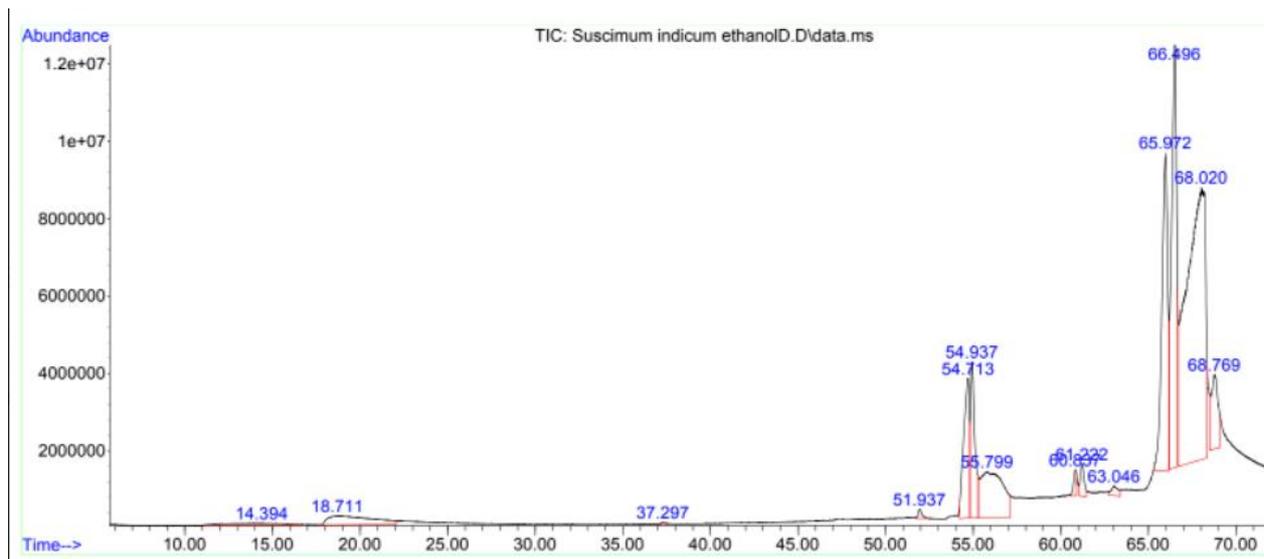
Fig 14.2 Standard

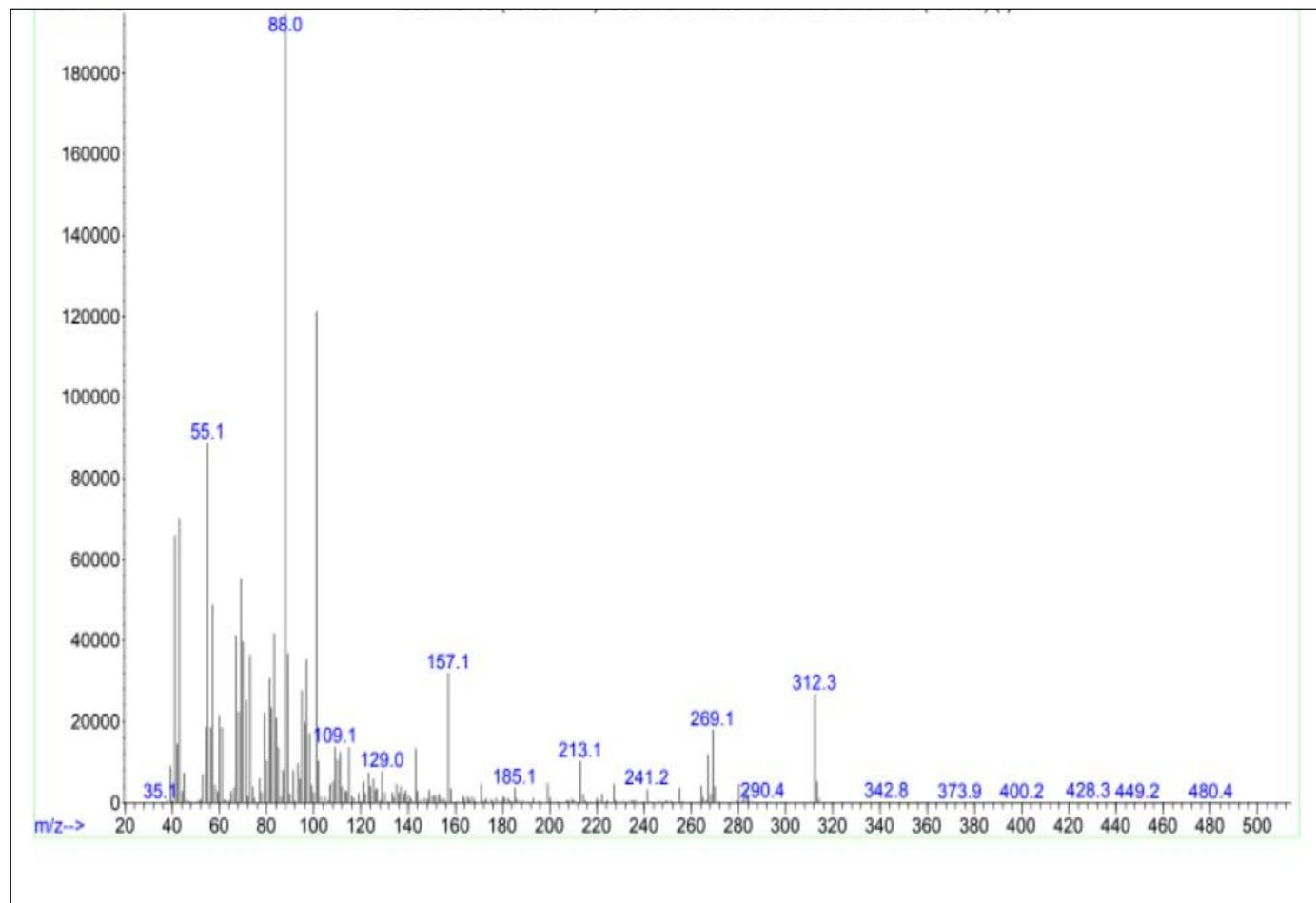
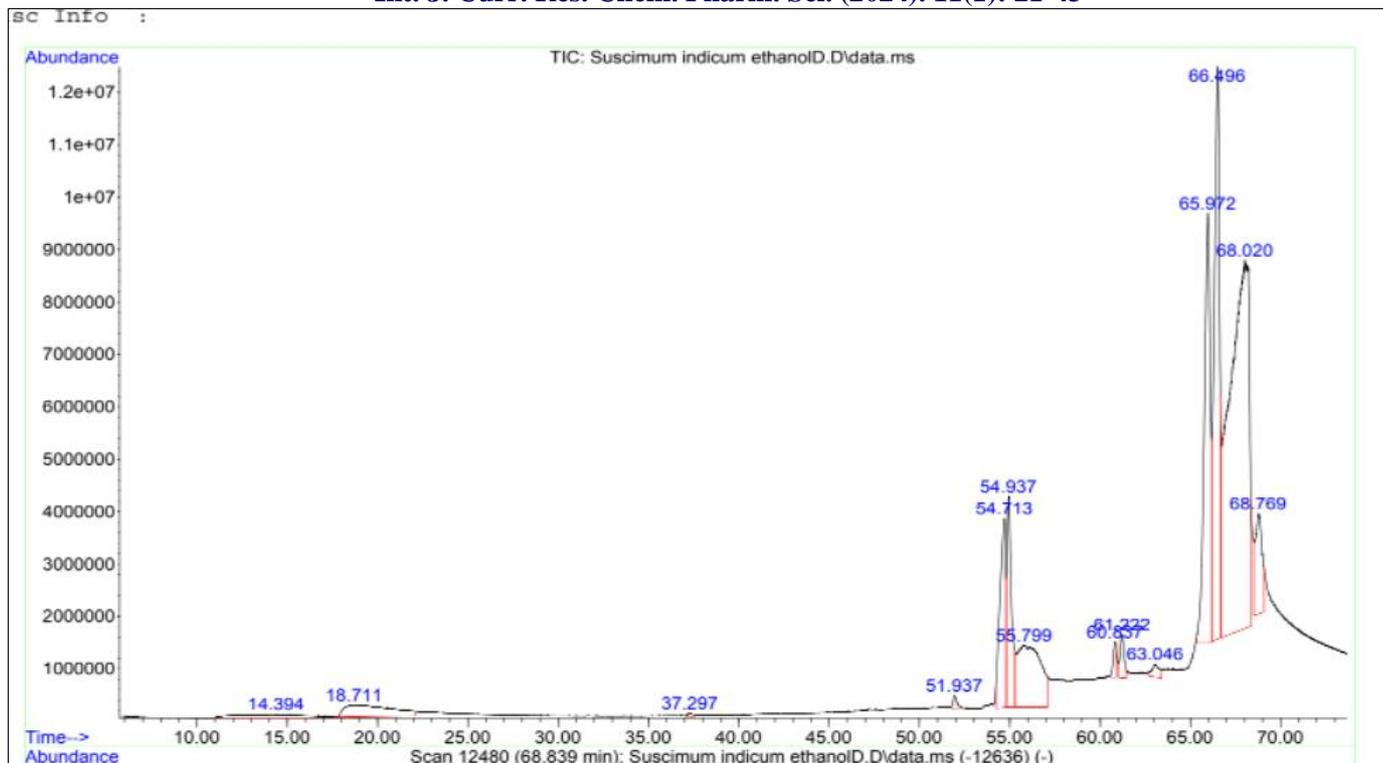
A doubly unsaturated fatty acid, occurring widely in plant glycosides. It is an essential fatty acid in mammalian nutrition and is used in the

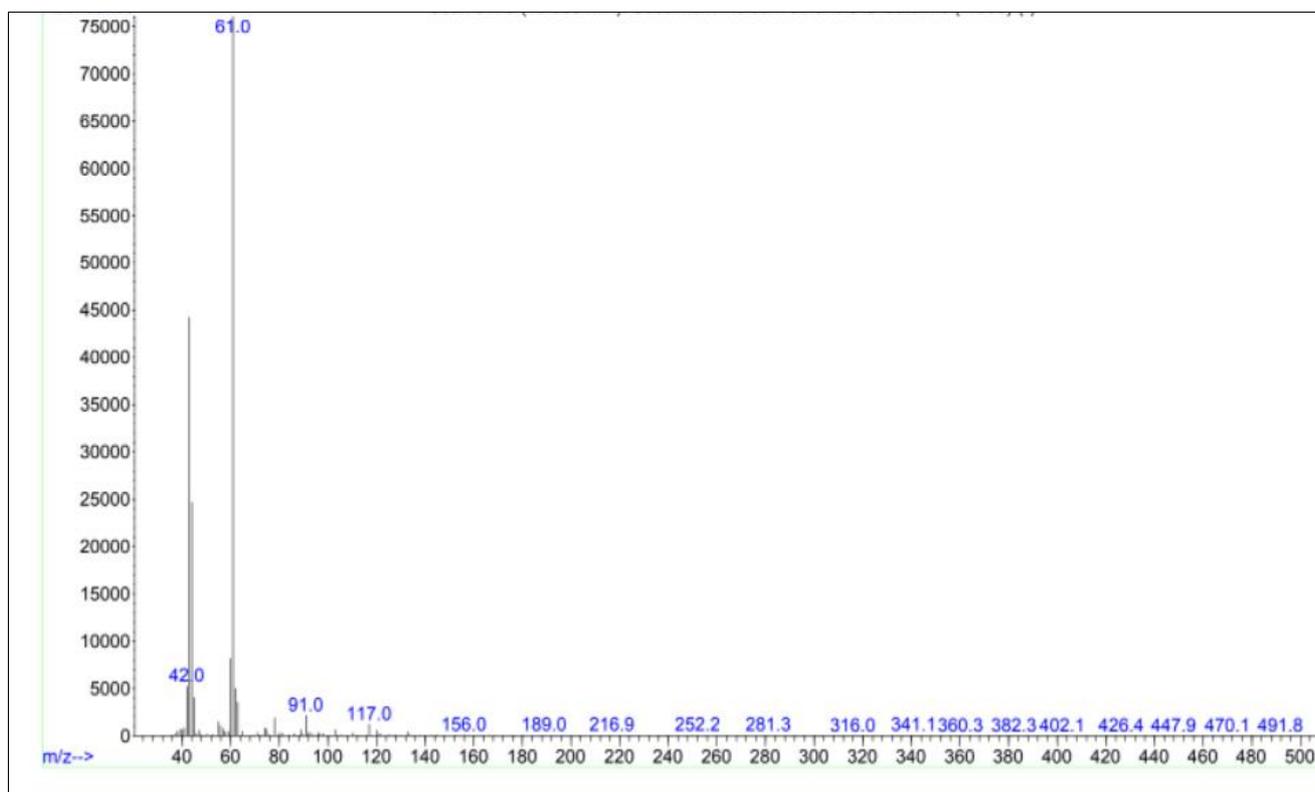
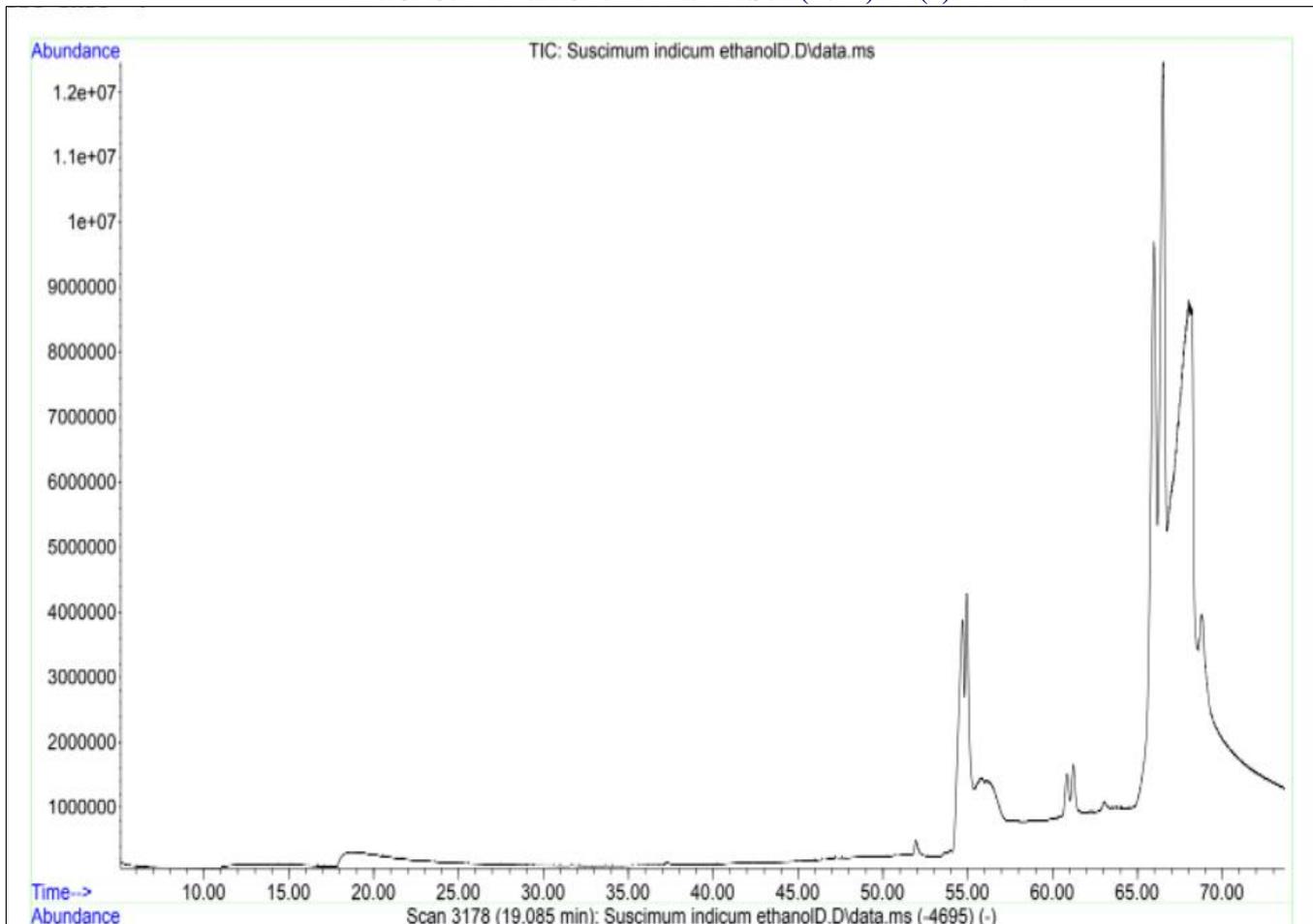
biosynthesis of prostaglandins and cell membranes.

**Data File:** *Suscimum indicum* ethanol D.D

**Sample:** *Suscimum indicum*\_Ethanol extract







The EESI extract's GCMS values are displayed as excellent oil components in the final analysis results.

## Conclusion

The results of present investigation show that the seed extract of *Sesamum indicum* (L). possess various phytonutrients, amino acids, and excellent antioxidant activity. The flavonoids, alkaloids, saponins, triterpenoids, anthroquinones, quione, tannins and steroids. The components present in GCMS is Palmitic acid, linolenic acid, Methyl esterate, Stearic acid, Glycerin, Oleic acid.

Medicinal plants constitute the basis of health care system in many societies. The recovery of the knowledge and the practices associated with these plant resources are a part of an important strategy in the discovery of new medicines and bettering the quality of life of poor rural communities.

Traditional medical knowledge of medicinal plants is not only useful for conservation of cultural traditions but also for community healthcare and drug development in the present and future. This traditional knowledge on the indigenous uses of the medicinal plants could boost new medicinal trails.

Herbal medicines that are rich in antioxidants and nutrients possibly be used to improve well-being and quality of life of person. *Sesamum indicum* seed extract are rich in nutrients and contains various compounds. Phenolic compounds like flavonoids, steroids, and tannins are the major source of antioxidants, which are highly present in *Sesamum indicum* seed. Antioxidant activity of the phenols is mainly due to their redox, hydrogen donor and singlet oxygen quenching ability. The phenolic compounds were highly correlated with antioxidant ability. Hence the synergistic activity of all the compounds in the extract might be used as a potential therapy.

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