# Analysis of therapeutic value of *Tinospora cordifolia*

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**Abstract -** *Medicinal plants are great sources of many chemicals used in the pharma industry. Crude plant extracts are mostly used in case of herbal medicines as these extracts comprise of mixtures of different phytochemical constituents or secondary metabolites with high medicinal value. In this study, the identification and interpretation of medicinally important phytoactive compounds of Tinospora cordifolia stem is done using the Gas Chromatography Mass Spectrometry (GCMS) technique. The study mainly focuses on the determination of the phytoconstituents present in the methanolic extract of stem of Tinospora cordifolia using GCMS. Mass spectrum of each of the phytoconstituents that occur in the methanolic extract of stem of Tinospora cordifolia was matched with NIST-14 and Willey-8 spectra libraries. A total of 82 peaks were found which corresponds to quite a high number of medicinally important phytocompounds in a plant part. It can be concluded as a result of this study that we can use Tinospora cordifolia as a plant based natural alternative for the current medicinal system. Gas Chromatography Mass Spectrometry (GCMS) analysis helps to postulate the formula and structure of phytoconstituents that can be utilized in the drug designing process and further research would help in developing novel drugs. Some of the very important phytoactive compounds like Squalene, Eicosalone, Alpha tocopherol, Stigmasterol, isopropyl-ester, Phytol, Pentanoic acid, Propyl ester or valproic acid, and 9, Linoelaidic acid, 3, 5-dimethoxy-acetophenone, Hexadecanoic acid, Methyl Ester, n-hexadecanoic acid, Neophytadiene, Lupeol were found to be present in the stem of Tinospora cordifolia as a result of GCMS Technique. Thus, the current study is basically focused on the evaluation of various phytoconstituents of Tinospora cordifolia justifying the utilization of this herb to treat various diseases. These findings support the traditional usage of Tinospora cordifolia in treatment of various diseases and disorders*

**Keywords:** *Bioactive components, GCMS, Tinospora cordifolia.*

## INTRODUCTION

Medicinal plants are of immense interest to the researchers in the field of biotechnology and pharmacology, as maximum drug based industries are dependent on medicinal plants for the production of medicinal compounds.

Plants are capable of producing enormous number of low molecular weight organic compounds commonly known as secondary metabolites,generally with unique as well as complex structures. Several such metabolites are known to possess interesting biological activities and have been used in pharmaceuticals. *Tinospora cordifolia* commonly known as Giloy in Hindi and Guduchi in Sanskrit is certainly one of the most important versatile herbs that is commonly found in India[1]. In addition to plains, the plant is found in the Himalayas as well, it can also be found at a height of about 1000 feet high and its occurrence is seen across a wide area in India that spreads from The Kumaon Mountains in the North to Kanyakumari in the South. It is also found in various countries in Asia like China, Sri Lanka, Thailand, Philippines, Malaysia as well as Africa. It has been given various National and International names like Gulancha in Bengali, Gulvel in Punjabi, Gilo in Oriya, Arab countries and Burma, Gulbel in China and Amritu in Marathi [1,2,3].

It is often cultivated as an ornamental plant and it can be easily propagated through stem cuttings. It can also be grown by sowing seeds in monsoon, but the growth of seedlings is found to be slower as compared to plants grown by cuttings [1, 4, and 5].

Giloy is a plant of therapeautic value that is perennial, deciduous climbing shrub with a succulent stem and alternate heart shaped leaves [1]. The stem of *Tinospora cordifolia* is generally found to be fleshy, succulent and climbing in nature. It has long filiform fleshy aerial roots. Its leaves are heart shaped, juicy and cordate.

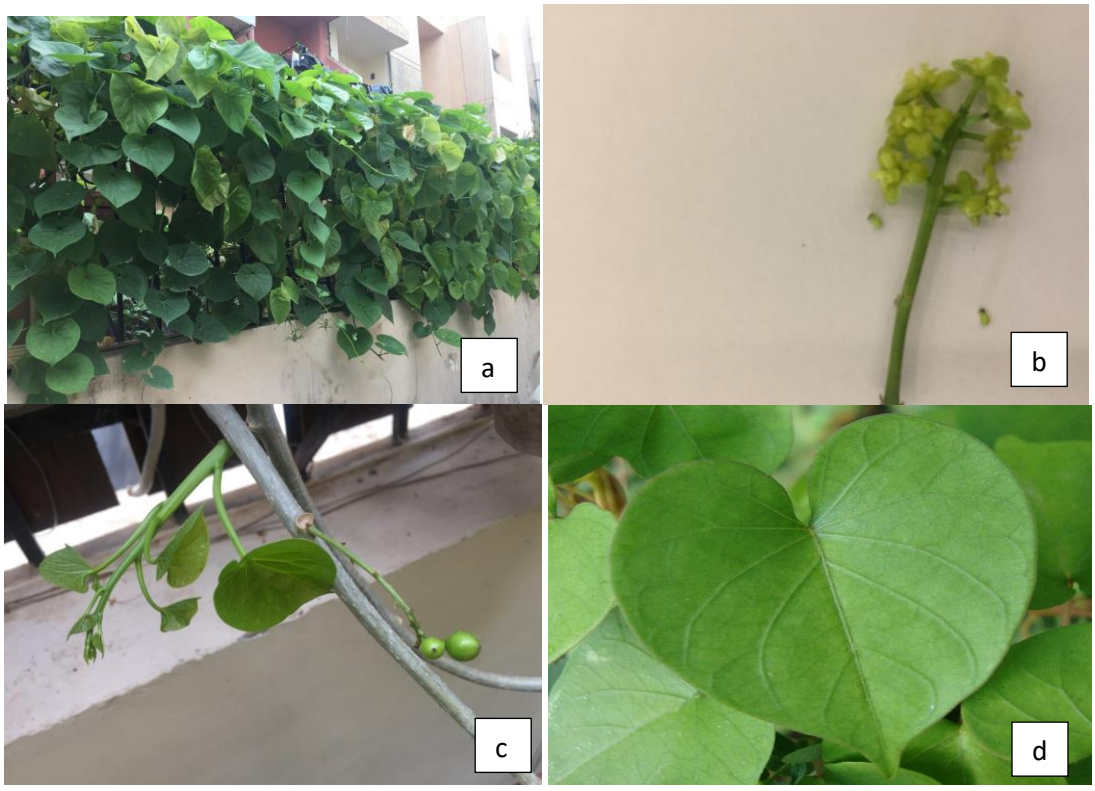


Figure 1(a) *Tinospora cordifolia* (Giloy); (b) Showing flower of *Tinospora cordifolia;* (c) showing *Tinospora cordifolia* fruit and leaves; (d) showing a healthy Leaf of *Tinospora cordifolia*

The lamina of leaf of *Tinospora cordifolia* is ovate, alternate or lobed, about 7- 9 nerved and membranous. The bark of *Tinospora cordifolia* is greyish or creamy white in colour with rosette like lenticels and is usually thin in nature [6, 7, 8]. The thread like, aerial and long filiform roots are found in *Tinospora cordifolia* and are seen usually arising from the branches [3, 8, 9]. Fruits of this plant appear shiny, orange reddish in colour and develop during winter season. Fruits appear pea shaped, fleshy, in aggregates of 1-3 ovoid, smooth drupelets and become red when fully mature. Seeds present in *Tinospora cordifolia* appear curved in shape; therefore this family is named as moonseed family [3, 8, 9 ]. Its flowers bloom in summer. Flowers of *Tinospora cordifolia* are unisexual and smaller in size. Male flowers are often found to occur in clusters while female flowers are seen as solitary. There are six sepals that are arranged in two whorls and are yellowish green in colour [10].

*Tinospora cordifolia* has been recognized to be a popular traditional medicinal plant and many researchers have found it to be an interesting source for various studies. Out of many reported medicinal properties few like hepatoprotective, immunomodulatory, anti-inflammatory and anti-neoplastic activities are important ones. This plant is designated as Rasayana and forms an important drug of the Ayurvedic Medicine. Almost all parts of this plant are reported to have various ethnobotanical and therapeutic uses. Giloy plants are specifically made to grow on other trees like neem tree as it can support the climber well.The traditional medicinal system will continue to be an important part of the health care sector since over 80 % of the population in the third world country relies on the use of traditional medicine. According to the World Health Organisation, Medicinal plants are quite prone to extinction, Therefore, for the purpose of scientific validation of traditional medicinal plants or the discovery of bioactive compounds for use as therapeutic drugs, the active principals in therapeautic plants needs to be identified.

In past decade, GCMS has become firmly established as a key technological platform to study the phytochemical profiling of secondary metabolites in plant species. GCMS is a method which combines the features of gas-liquid chromatography and mass spectrometry for the identification of various


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phytoconstituents within the test sample.In recent years GCMS analysis has been extensively used for the determination of non polar components,and volatile essential oil,fatty acids,lipids,terpenoids,steroids and alkaloids.

*Tinospora cordifolia* stem extracts were prepared in methanolic solvent to study the phytochemical profile using Gas Chromatography Mass Spectrometry technique. GCMS analysis of stem extract of Tinospora cordifolia revealed the occurrence of some of the major peaks like Lupeol (RT: 52.68),alpha-tocopherol(RT:46.361 ), Pytol (RT:32.026), Squalene (RT:27.323 ),Hexadecanoic acid(RT:29.086), etc. A number of secondary metabolites like Linolaidic acid, Gamma-sitosterol, Octadecanoic Acid and Phytol were found as a result of GCMS Analysis of *Tinospora cordifolia* stem extract. These compounds are some of the examples and are discussed in detail in Results section with references. This study can be used further as a referential source of valuable information related to amount and number of phytoconstituents present in *Tinospora cordifolia* that can help in the formulation of novel drugs.

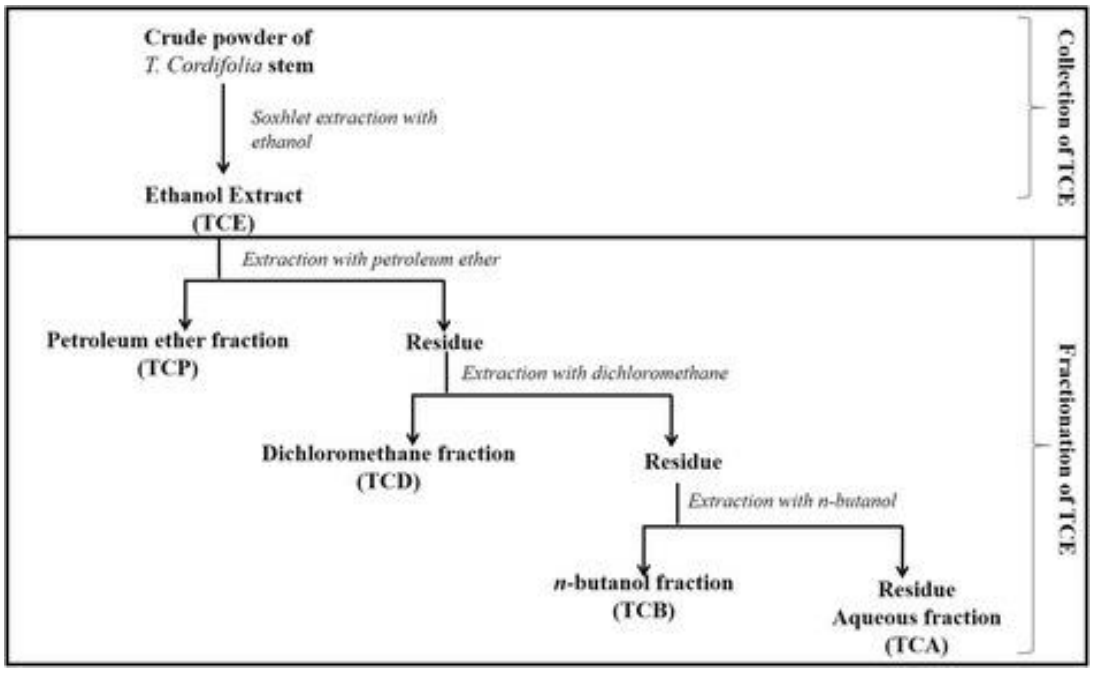
## METHODS

### Collection of Plant Material (Stem of *Tinospora cordifolia*):

The Stem part of *Tinospora cordifolia* was collected from Botanical Garden, JNU Campus, New Delhi. For the identification purpose, a voucher specimen and a Herbarium were deposited and voucher number (Acc.No.118379) was obtained from The Botanical Survey of India, The North Regional Centre located at Dehradun.

### Preparation of extract for phytochemical analysis of *Tinospora cordifolia* stem:

The collected part of plant (*Tinospora cordifolia)* like the stem in this case was dried in shade. Then after that powdered form of stems of *Tinospora cordifolia* were extracted with absolute ethanol by using soxhlet extraction method at 60 °C. After the extraction procedure, the extract was concentrated and kept in a desiccator under reduced pressure and controlled temperature. Ethanolic extract of *Tinospora cordifolia* was dissolved in water and partitioning was carried out three times each with petroleum ether, Dichloromethane, *n*-butanol and aqueous and was further analysed. All fractions were evaporated by rotary evaporator to dryness under decreased pressure and controlled temperature and were stored for further utilization. Later on its percentage yield was determined and noted.



**Qualitative Analysis of various components present in stem extract *of Tinospora cordifolia***:

The Ethanolic and aqueous stem extracts of *Tinospora cordifolia* were analyzed to show the occurrence of various compounds in it by the using certain methods.

*1. Tests for the presence of Flavonoids*- The test for Flavonoids was done by adding few pieces of magnesium ribbon and HCl concentrated in aqueous plant extract, then after couple of minutes, pink color appears and thus confirms the presence of flavonoid.

*2. Test to check the presence of Anthraquinones*- The test for Anthraquinones was done by adding 10ml of benzene to 6 g of stem extract of *Tinospora cordifolia* and then, it was further soaked for 10 min before being filtered. Further added 10ml of 10% ammonia in the filtrate followed by shaking vigorously. Appearance of pinkish color showed Anthraquinones in solution.

*3. Test to check the presence of Tannins*- The test to check the presence of Tannins was seen on the addition of 10ml of bromine water in 0.5 g of *Tinospora cordifolia* stem aqueous extract and then the decoloration of bromine water proved that the tannins were present.

*4. Test for the presence of Steroids*- The test for steroids was done by adding 2ml of chloroform and conc. $H_2SO_4$ with 5ml aqueous *Tinospora cordifolia* stem extract and then the appearance of red color in lower layer indicated the presence of steroids.

*5. Test to check the presence of Saponins*- The test for saponins was done by the addition of recommended amount of autoclaved distilled water into the aqueous stem extract of *Tinospora cordifolia* and further mixed properly. Olive oil was added to the resulting froth and the appearance of foam indicated the presence of saponins.

*6. Test for presence of Terpenoids*- The test for terpenoids was done by adding 2 ml of chloroform with 5 ml of aqueous extract of *Tinospora cordifolia* stem and the resulting solution was evaporated on the water bath and boiled with 3 ml of conc. $H_2SO_4$. Appearance of grey color indicated the presence of Terpenoids.

*7. Tests to check the presence of Glycosides*- The test for glycosides was done by performing Liebermann's Test and Salkowski's Test.

**Gas Chromatography Mass Spectrometry (GCMS) ANALYSIS:**

**Collection of Plant Material (Stem of *Tinospora cordifolia*):**

The Stem part of *Tinospora cordifolia* was collected from Botanical Garden, JNU Campus, New Delhi. For the identification purpose, a voucher specimen and a Herbarium were deposited and voucher number (Acc.No.118379) was obtained from The Botanical Survey of India, The North Regional Centre located at Dehradun

**Extraction of Plant Material:**

**Preparation of stem extract of *Tinospora cordifolia* for Gas Chromatography Mass Spectroscopy (GCMS) analysis:** The plant part (thick fleshy stem) was cleaned and dried and then further it was powdered with the help of mixer grinder. About 15 g of dried stem powder of *Tinospora cordifolia* was taken and soaked in 150 ml methanol. With constant shaking, it was placed for 72 hrs at normal room temperature. After incubation, solutions were filtered properly and its filtrate was kept at room temperature for drying. After drying, the weight of extract was measured and according to weight, solvent was further added and maintained the concentration of the plant extract as 25 mg/ml. Further storage was done by keeping it at a temperature of 4ºC in air tight containers for future usage. It was then taken to AIRF (Advanced Instrumentation Research Facility), Jawaharlal Nehru University (JNU), Delhi, India for further determination and examination of its phytoconstituents.

**Preparation of Stock Solutions:** The *Tinospora cordifolia* stem extracts were reconstituted in methanol and then methanolic extracts (2 µl) were injected for further analysis using the GCMS Technique.

**Gas Chromatography Mass Spectrometry (GCMS) Technique**: Gas Chromatograph Mass Spectrometer (GCMS) - Shimadzu QP-2010 Plus with Thermal Desorption System TD 20 was used to perform the analysis of the plant extract at the AIRF (Advanced Instrumentation Research Facility) , JNU Campus, New Delhi.

The procedure for GCMS analysis was performed using the **GCMS-QP2010 Ultra SHIMADZU** (S.No.74707) with GCMS solution software at high resolution. This particular instrument is equipped with a column i.e. **Rxi-5Sil MS Column**, which is fused with silica capillary column of size 30m x 0.25mmID x 0.25 µmdf. To detect GCMS results, system was made



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to run having ionization energy of 70 eV. Helium gas (99.99%) was specifically used as a carrier gas and injection volume of 2 µl was used.

The temperature of injector was 270 °C and the ion-source temperature was maintained at 230 °C. The interphase temperature was 280°C and the oven temperature was further programmed starting at 50 °C increasing by 6 °C/min to 250 °C, then after that the hold time for 2 minutes was maintained. After that it was made to run at the rate of 15°C/min to 280°C thus finally ending with the hold time of 20 minutes.

The mass spectra were recorded. The recorded cut time for solvent was 5 min, and the total GCMS running time was taken as 60.32 min. Average peak area to the total area was compared to find out the relative percentage amount of each phytoconstituent.

**Identification of Components**: Determination of the mass spectrum GCMS was performed using the already existing database of National Institute Standard and Technology (NIST) 14 and WILEY 8 libraries. Here in this case, the spectra of unknown phytoconstituents or the sample to be studied was compared with the spectra of the known compounds that have been stored in the NIST 14 and WILEY 8 libraries. Certain number of hits were observed and recorded. It is further used to determine the compound name along with its molecular formula and molecular weight and also other parameters like their structures and common names that can be studied later on using bioinformatics tools based on these observations. Some of the very important phytoactive compounds like Squalene, Eicosalone, Alpha tocopherol, Stigmasterol, isopropyl-ester, Phytol, Pentanoic acid, Propyl ester or valproic acid, Phytol and 9, Linoelaidic acid, 3, 5-dimethoxy-acetophenone, Hexadecanoic acid, Methyl Ester, n-hexadecanoic acid, Neophytadiene, Lupeol were found to be present in the stem of *Tinospora cordifolia* as a result of GCMS Technique.

## RESULTS AND DISCUSSION

**Result of Phytochemical Analysis:** As mentioned in Table no.1, results showed the presence of Carbohydrates, Tannins, Saponins, Triterpenoids, Flavonoids, Steroids, Glycosides and Alkaloids in the Ethanolic stem extract while Quinones were found to be absent in both Aqueous and Ethanolic Stem extracts. Also it was observed that Saponins and Steroids were not found in Aqueous Extracts of stem of *Tinospora cordifolia*.

**Table 1. Showing the presence or absence of various phytoconstituents in *Tinospora cordifolia***

| Plant Constituent | Aqueous extract (Stem) | Ethanolic Extract (Stem) |
|---|---|---|
| Carbohydrates | + | + |
| Tannins | + | + |
| Saponins | - | + |
| Triterpenoids | + | + |
| Flavonoids | + | + |
| Quinones | - | - |
| Steroids | - | + |
| Glycosides | + | + |
| Alkaloids | + | + |

*Phytochemical test: '+'- Present and '–' - Absent*

**GCMS Technique Result:** The results of GCMS analysis of *Tinospora cordifolia* were found to be quite interesting. The results of the study of methanolic *Tinospora cordifolia* stem extracts indicated the appearance of eighty four peaks. Various medicinally important compounds were identified here in *Tinospora cordifolia* stem extract by the utilization of GCMS Technique. The table given below shows some of the important bioactive compounds found in the stem extract of *Tinospora cordifolia*.

The given results of GCMS technique in Table 2(see appendix) shows various peaks along with their retention time and area percentage of compounds found in stem of *Tinospora cordifolia*. Various medicinally important compounds like alpha tocopherol, squalene, palmitin, stigmasterol etc. are seen to be present in quite higher amounts in the stem of Giloy.

Various kinds of secondary metabolites were found as a result of GCMS Analysis of *Tinospora cordifolia* stem extract. Compounds like Linolaidic acid, Gamma-sitosterol, Octadecanoic Acid and Phytol have shown higher peaks in the graphs with their retention time as 32.395, 32.823 and 32.026, respectively. Hexadecanoic acid is also found to show higher peak value with a retention time of 29.712. The area percentage was found to be 13.56 for Tetradecenal, 11.93 for Hexadecanoic acid and 8.79 for Linolaidic acid. Gamma- sitosterol shows area percentage of 9.19 and retention time of 50.653. Hexadecanoic acid was found to show anti-oxidant properties as reported in previous studies. Literature studies also reports anti-inflammatory activity of Tetra-decenal as well.Some of the important bioactive compounds found in Table 2 are of great medicinal value and their fragmentation patterns along with their molecular structures have been studied here (Fig 3-Fig 20).

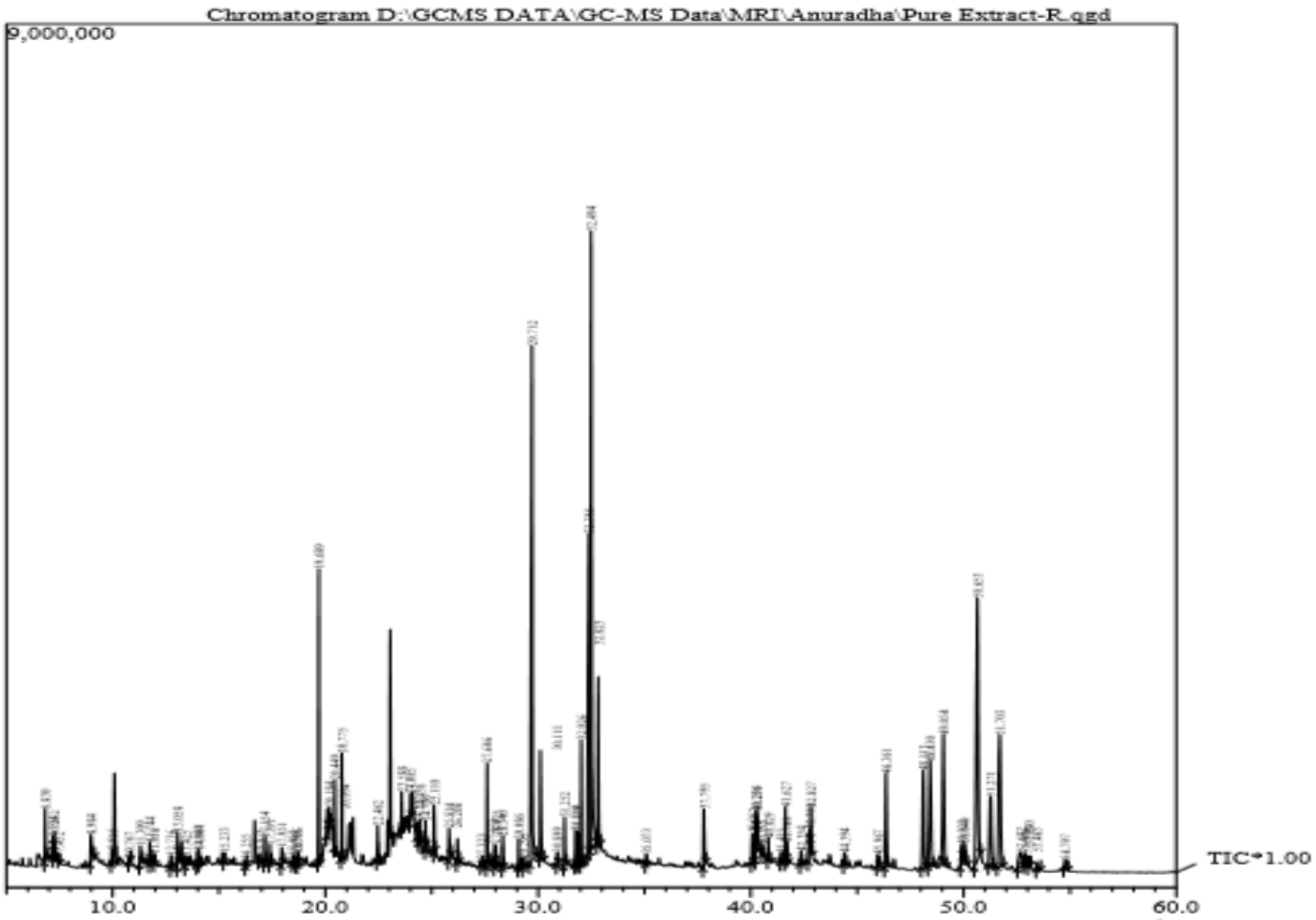


**Figure 2: GCMS result of methanolic extract of *Tinospora cordifolia***

Fragmentation patterns of some important compounds found in *Tinospora cordifolia* are given below. Fragmentation pattern holds importance as it gives detailed information about the compounds observed in spectra analysis. The phytoconstituents showing high retention time have been analyzed here using NIST and WILEY libraries. In GCMS, the m/z values on the x axis represents mass to charge ratio where m stands for mass and z stands for charged number of ions and y axis represent the relative abundance. The mass spectrum is represented as a stick diagram where the tallest peak in the stick diagram is called the base peak. Molecular structures of the compounds identified in Tinospora have been given with their fragmentation patterns in the following figures.

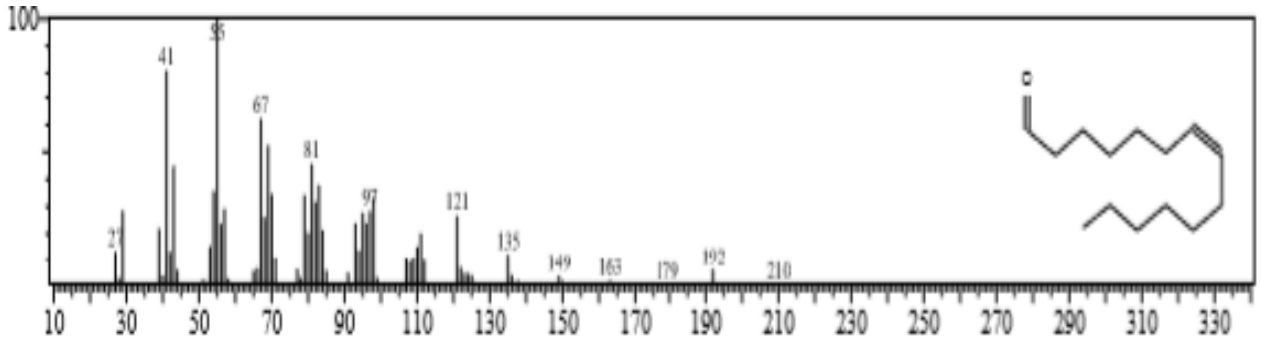


Fig 3**.** Fragmentation pattern of Z-7-Tetradecenal showing its molecular structure.


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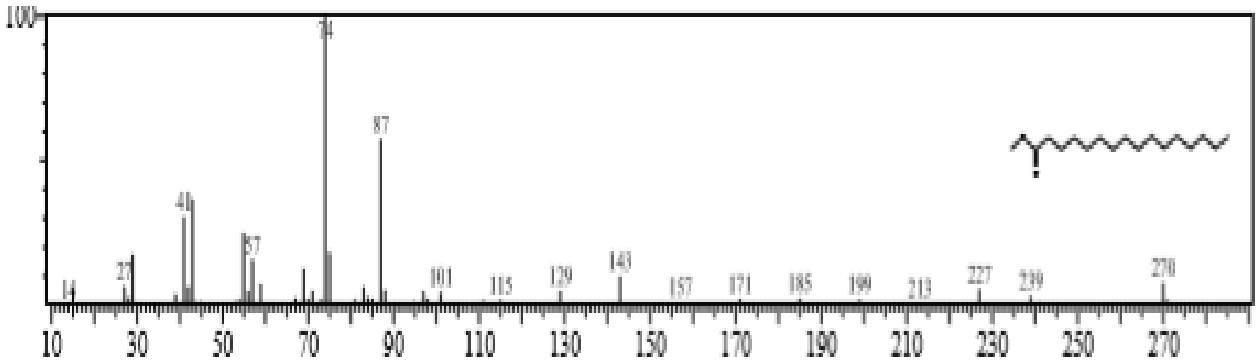

Fig 4. Fragmentation pattern of n-Hexadecanoic acid showing its molecular structure.

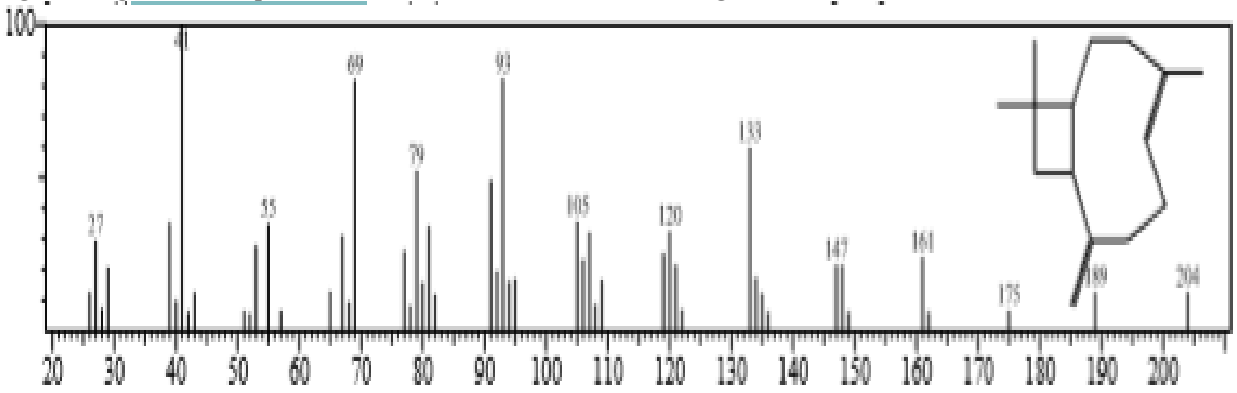

Fig 5. Fragmentation pattern of Beta caryophyllen showing its molecular structure.

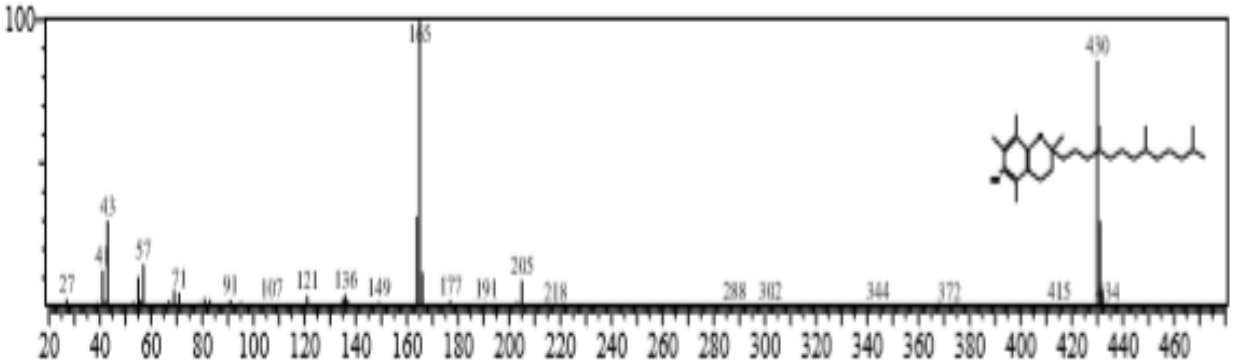

Fig 6. Fragmentation pattern of dl-.alpha.-Tocopherol showing its molecular structure.

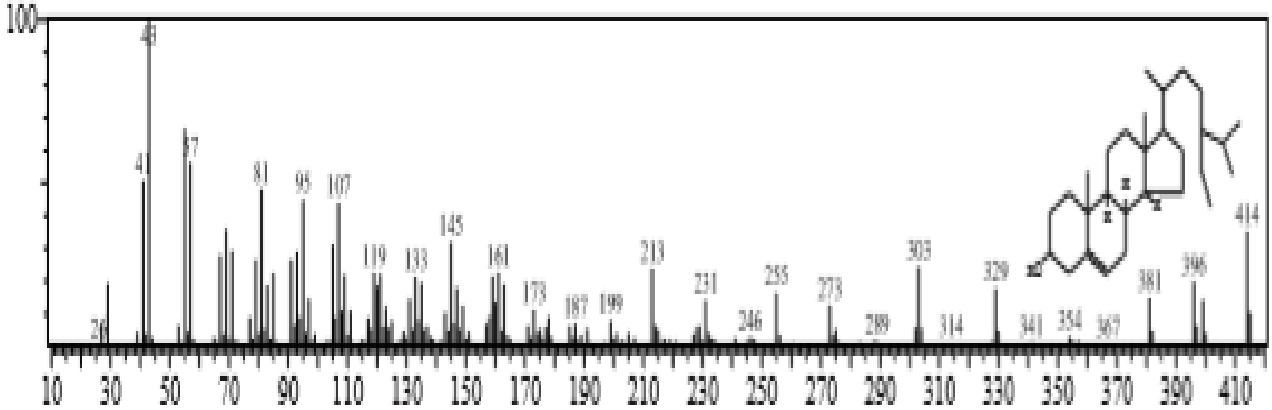

Fig 7. Fragmentation pattern of gamma.-Sitosterol showing its structure.

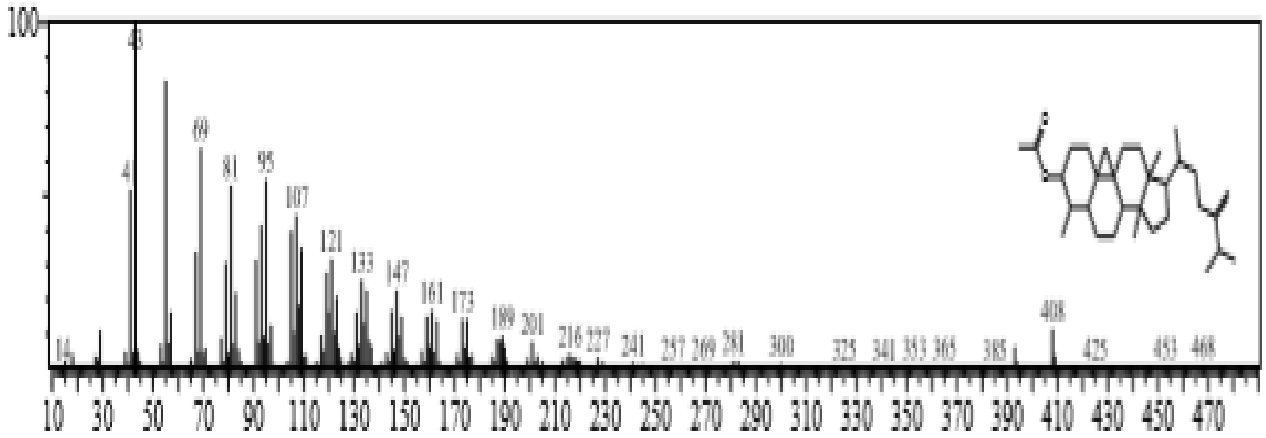


Fig 8. Fragmentation pattern of Cycloeucalenyl acetate showing its molecular structure

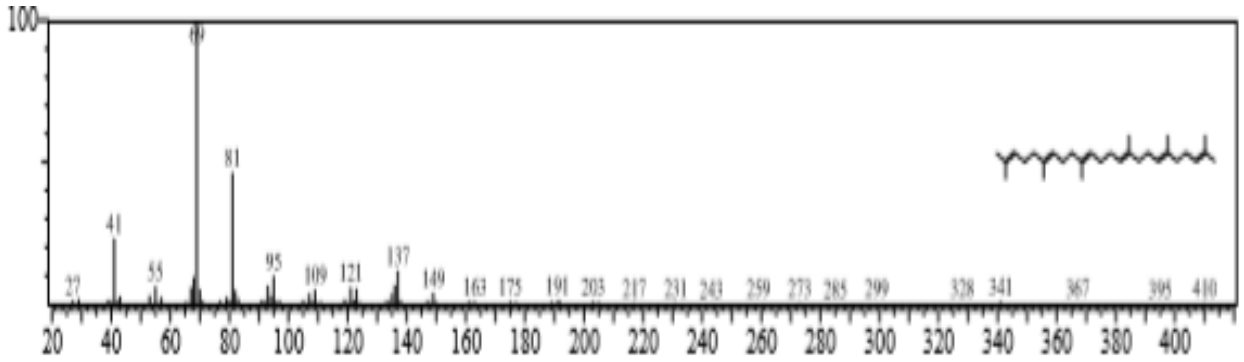


Fig 9. Fragmentation pattern of Supraene showing its molecular structure

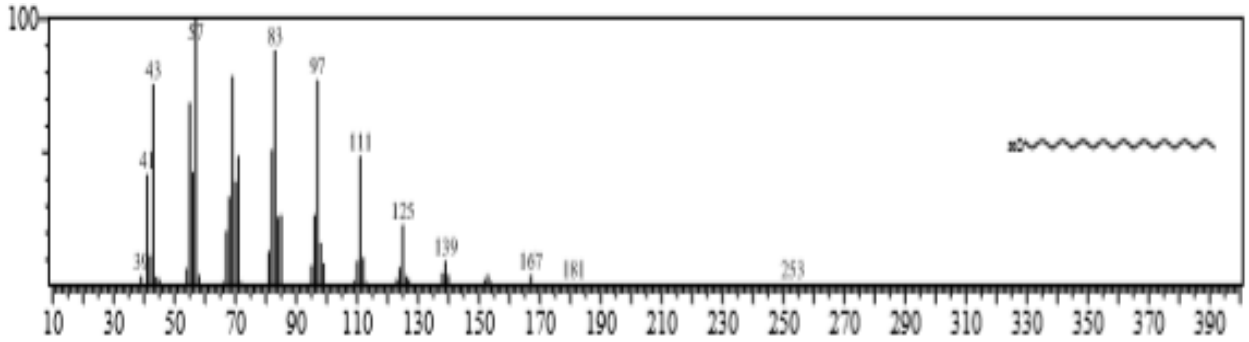


Fig.10. Fragmentation pattern of 1-ICOSANOLshowing its molecular structure

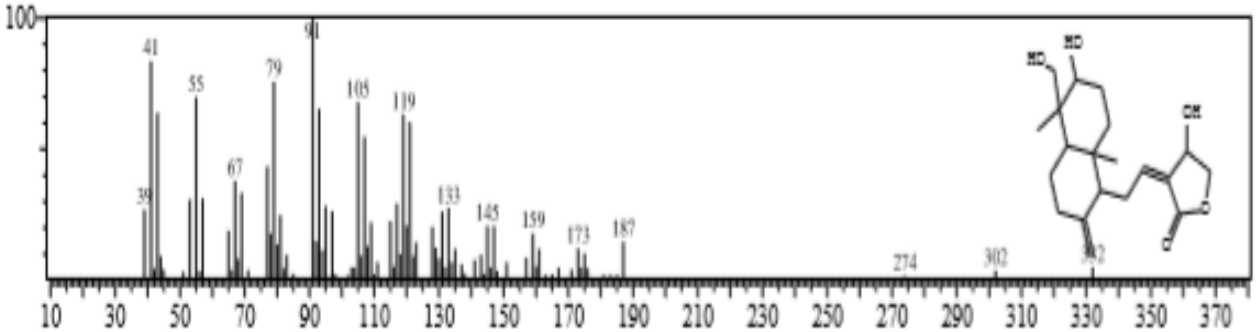


Fig.11. Fragmentation pattern of andrographolide showing its molecular structure .

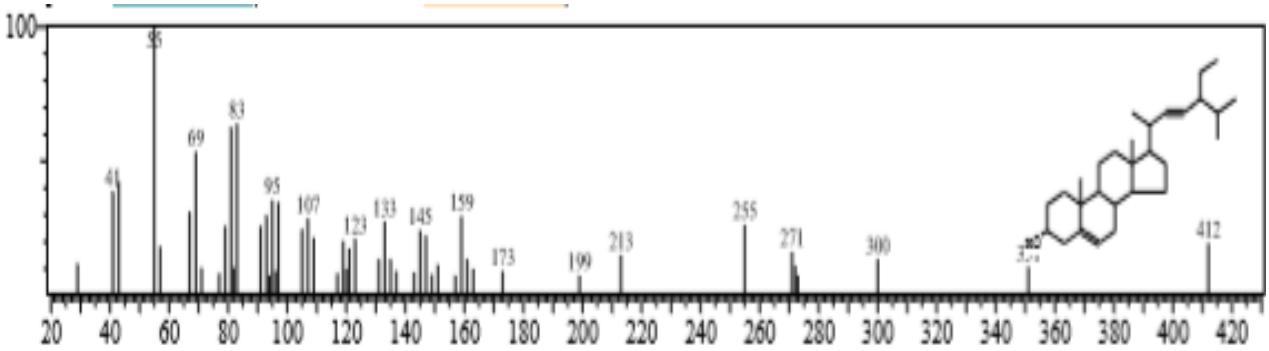


Fig.12. Fragmentation pattern ofSTIGMASTA-5 (stigmasterol) showing its molecular structure.

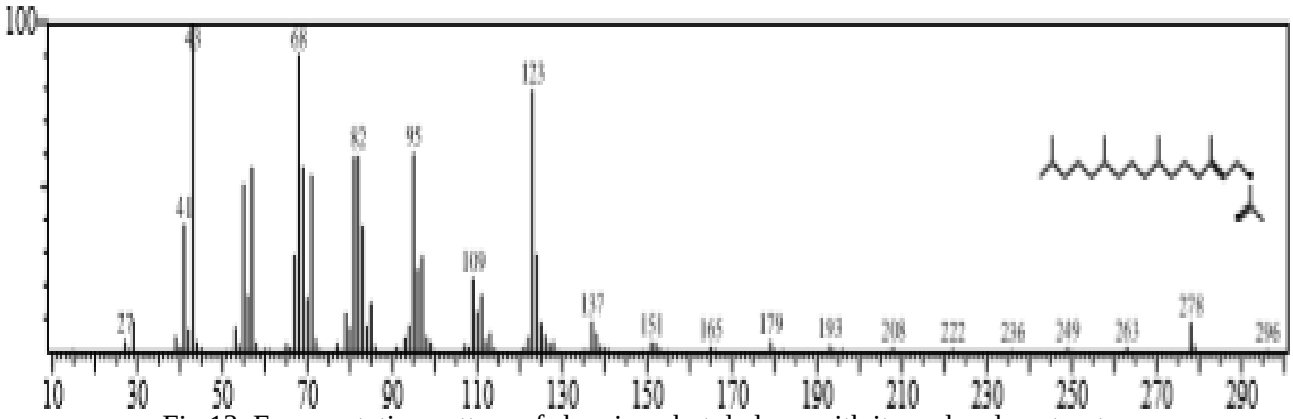


Fig.13. Fragmentation pattern of showing phytol along with its molecular structure .

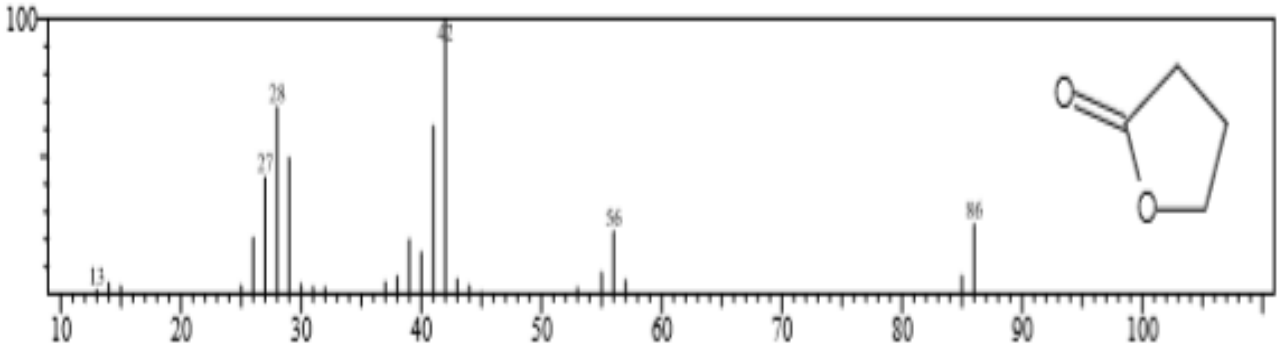


Fig.14.Fragmentation pattern of showing Butyrolactone along with its molecular structure .

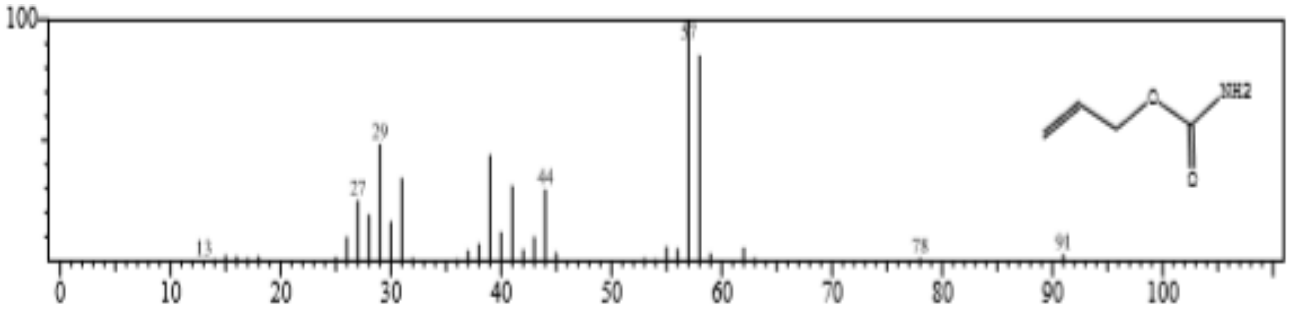


Fig.15. Fragmentation pattern of showing Allylcarbamate along with its molecular structure .

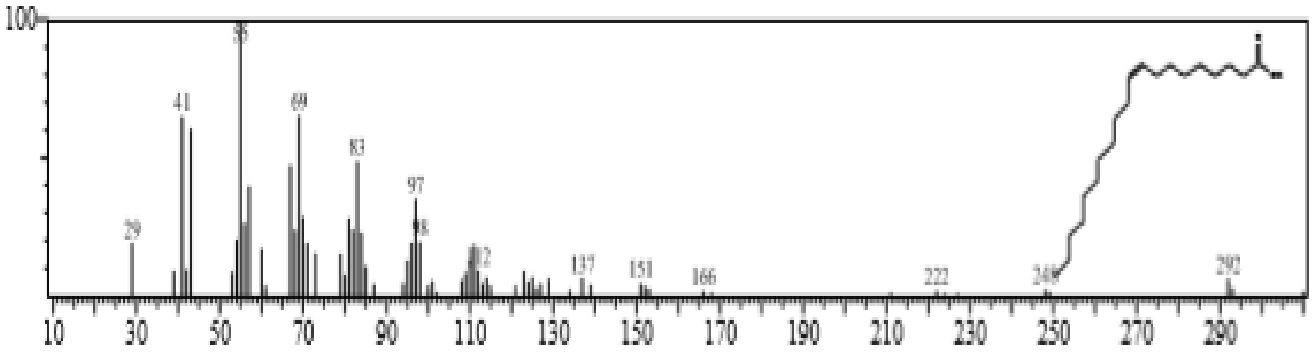


Fig.16.Fragmentation pattern of showing9-Eicosenoic acid along with its molecular structure .

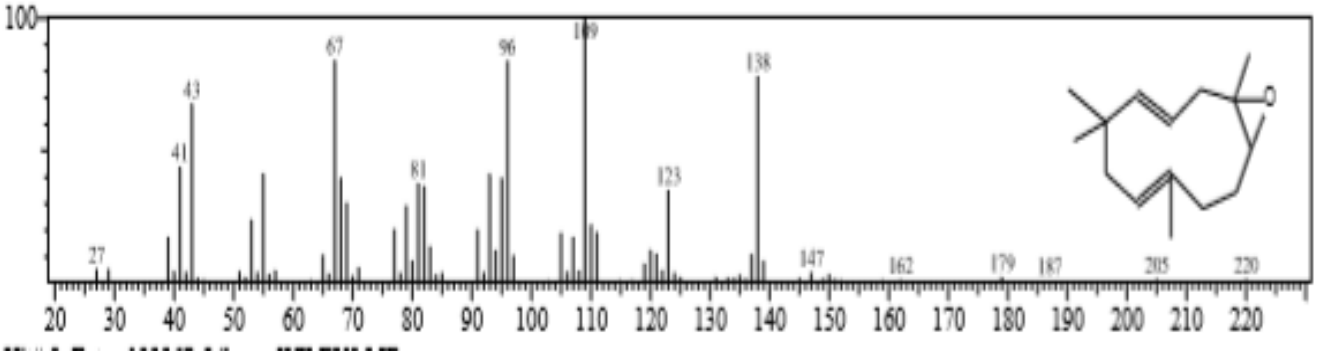


Fig.17.Fragmentation pattern of showing Humuladienone along with its molecular structure .

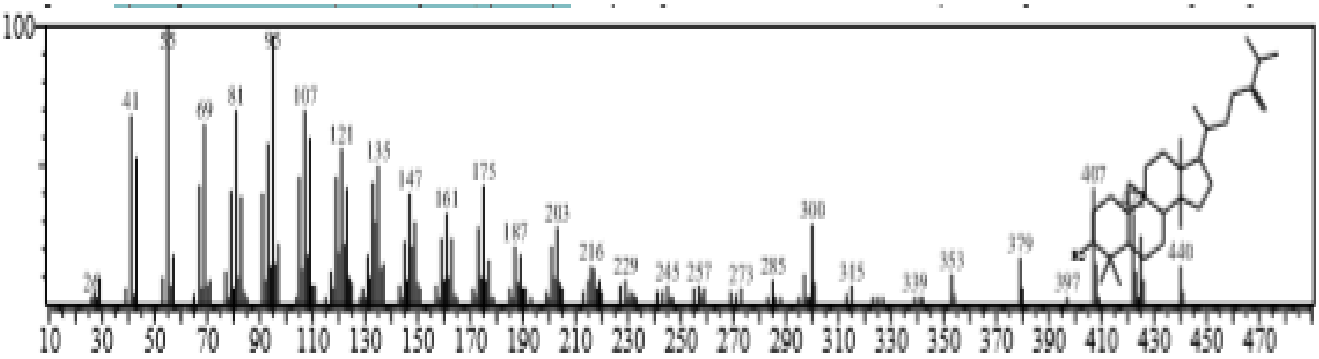


Fig.18. Fragmentation pattern of showing919-Cyclolanostan-3-ol, 24-methylene-, (3.beta.)-along with its molecular structure .

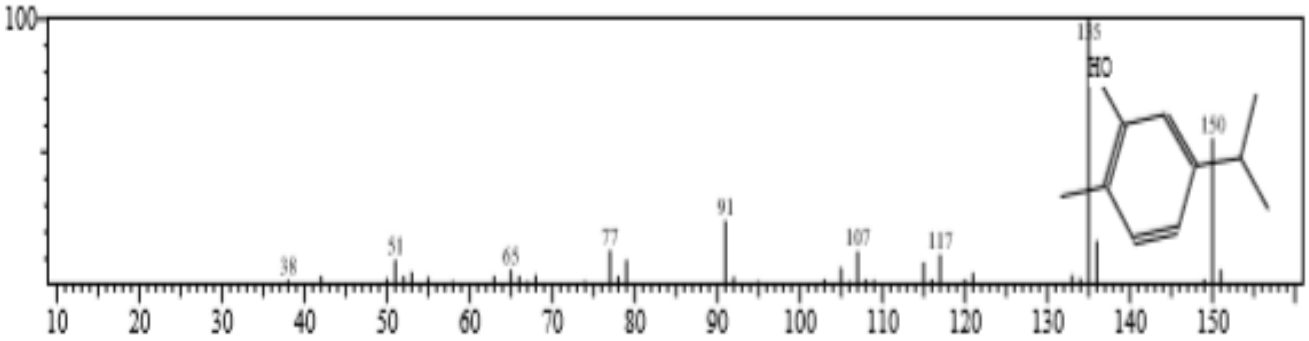


Fig.19.Fragmentation pattern of showing5-ISOPROPYL-2-METHYLPHENOLalong with its molecular structure .

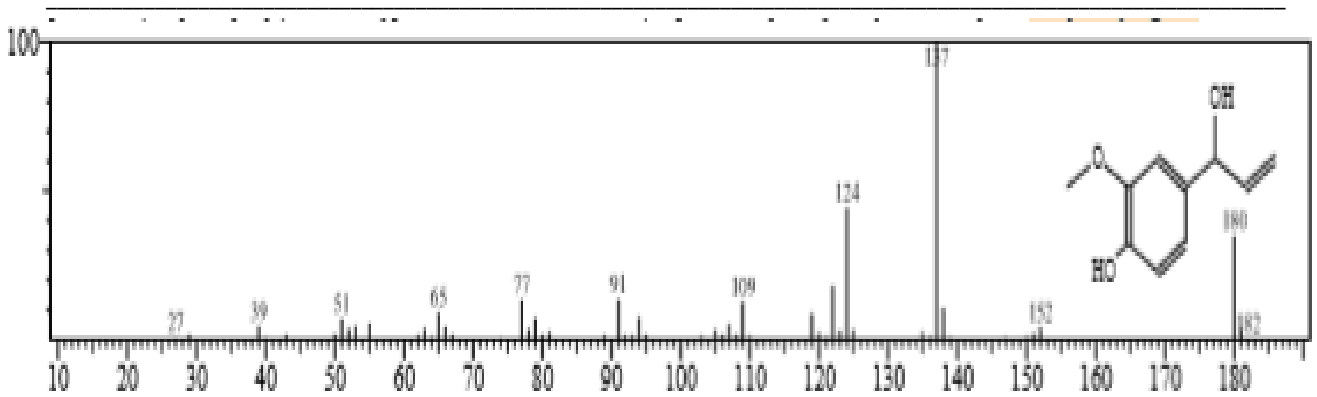


Fig.20.Fragmentation pattern of showing1'-Hydroxyeugenol along with its molecular structure.

**Table 3 - Some medicinally important phytocompounds found in *Tinospora cordifolia***

| S No. | Retention time of major peaks | Matched Compound name | Mol wt. | Formula | Uses/Medicinal properties |
|---|---|---|---|---|---|
| 1. | 32.4 | Z-7-Tetradecenal | 210 | C14H26O | Anti-inflammatory activity [11] |
| 2. | 29.71 | N -Hexadecanoic acid | 256 | C16H32O2 | Antioxidant [8] |
| 3. | 19.6 | Beta-Caryophyllen | 204 | C15H24 | Anti-Carcinogenic [7,8] |
| 4. | 50.6 | Clionasterol | 414 | C29H50O | Anticancer and Antioxidant [13] |
| 5. | 51.7 | Lupeol | 426 | C30H50O | Antioxidant and Antibacterial activity [13] |
| 6. | 41.6 | Squalene | 410 | C30H50 | Adjunctive Cancer therapy, provides skin protection [14] |
| 7. | 46.3 | dl-alpha-Tocopherol | 430 | C29H50O2 | Potent Antioxidant and cytoprotective, hepatoprotective [14,15] |
| 8. | 45.9 | 1-Icosanol | 298 | C20H42O | Used to cure malaria and filariasis[16] |
| 9. | 24.3 | Andrographolide | 350 | C20H30O5 | Hepatoprotective[17] |
| 10. | 49.0 | Stigmasterol | 412 | C29H48O | Anticancer and inhibit tumor promotion, [18] |
| 11. | 20.7 | Isopropylester | 168 | C10H16O2 | Antioxidant and Antiageing properties [20] |
| 12. | 27.6 | Phytol | 338 | C22H42O2 | Antioxidant [8] |
| 13. | 6.8 | Butyrolactone | 86 | C4H6O2 | To Treat Rheumatoid arithritis [21] |
| 14. | 8.9 | Allyl carbamate | 101 | C4H7NO2 | |
| 15. | 30.8 | 9-Eicosenoic acid, | 310 | C20H38O2 | Diabetes [22] |
| 16. | 23.5 | Humuladienone | 220 | C15H24O | Anticancer properties [8] |
| 17. | 54.7 | 24-methylenecycloartenol | 440 | C31H52O | Antioxidant [8] |
| 18. | 22.4 | 2', 4'-Dimethoxyacetophenone | 180 | C10H12O3 | Antioxidant [8] |
| 19. | 52.9 | Handianol | 426.7 | C30H50O | Anti-inflammatory[25] |
| 20. | 35.0 | Carvacrol | 150 | C10H14O | Anti- depressant [24] |
| 21. | 25.8 | 1'-Hydroxyeugenol | 180 | C10H12O3 | Anti-inflammatory activity [11] |

As shown in Table 3, the major components present in stem of *Tinospora cordifolia* were Lupeol (RT:51), Clionasterol (RT:50), Alphatocopherol (RT:46.3), Icosanol (RT:45), Squalene (RT:41),Carvacrol (RT:35) and Phytol (RT:27). According to the above given Table3 , Handianol (RT:52.9) ,Hydroxyeugenol (RT: 25.8) and Tetradecenal (RT: 32.4) shows anti-inflammatory effects.These are some of the major Phytochemical constituents which contribute to the medicinal activity of this plant. The stem contains Dimethoxyacetophenone, dl-alpha-Tocopherol and N - Hexadecanoic acid with high retention time periods that are considered mainly to be responsible for various antimicrobial properties. The presence of these compounds attributes to its antioxidative property and is also thought to be responsible for inhibiting the lipid peroxidation process. This property helps in maintaining good health and prevents the changes occurrence of heart diseases including many other biochemical diseases as oxidative stress is the hallmark of such diseases. Other compounds like Beta Caryophyllene, S, stigmasterol, Humuladienone etc. (Table 3) have been observed to possess anti-cancer properties.

## CONCLUSION

The phytochemical analysis showed that the methanolic extract of *Tinospora cordifolia* contain various phytoconstituents having immense potential bioactivity.Our study using GCMS technique revealed the presence of 82 phytoconstituents in this particular plant which is very interesting and makes it a target for further studies. Further research on the phytoconstituents present in this plant can lead to the treatment of many diseases by the development of novel drugs with lesser known side effects. Table 2 revealed the presence of various important phytoconstituents in *Tinospora cordifolia* along with their retention time and area percentage. Table 3 gives the information about the medicinal importance of various phytoconstituents found in *Tinospora cordifolia*.

The GCMS Technique used to study the methanolic extract of *Tinospora cordifolia* showed the presence of pharmacologically active phytocompounds that possess various medicinally important properties like antioxidants and anticancer compounds. Therefore, it is recommended as a plant of phytopharmaceutical importance and the quality of this herb can be improved through secondary metabolites production .However, isolation of these individual phytochemical constituents and subjecting it to pharmacological activity will definitely provide fruitful results. These identified phytoconstituents presumed to be responsible for medicinal properties shown by this plant. Thus it can be used as a source for developing novel drug formulations. The correlation between the phytoconstituents with their biological activities is now being a matter of novel thought process.

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## APPENDIX

**Table 2. Peak Report TIC**

| Peak# | R. Time | Area | Area% | Name |
|---|---|---|---|---|
| 1 | 6.830 | 1763958 | 0.94 | Butanoic acid, 4-hydroxy- |
| 2 | 7.194 | 283606 | 0.15 | 2-Cyclopenten-1-one, 2-hydroxy- |
| 3 | 7.252 | 437912 | 0.23 | -α-thujene |
| 4 | 7.452 | 190877 | 0.10 | α-Pinene |
| 5 | 8.984 | 1674531 | 0.89 | 2-Hydroxy-gamma-butyrolactone |
| 6 | 9.956 | 307897 | 0.16 | β-Cymene; |
| 7 | 10.767 | 119368 | 0.06 | 2,5-ANHYDRO-1,6-DIDEOXYHEXO-3,4-DIULOSE |

| | | | | |
|---|---|---|---|---|
| 9 | 11.744 | 607455 | 0.32 | CYCLOPROPYLMETHANOL |
| 10 | 12.736 | 411418 | 0.22 | 2-ACETYL-2-HYDROXY-.GAMMA.-BUTYROLACTO |
| 12 | 13.425 | 240894 | 0.13 | L-Menthone |
| 13 | 14.061 | 314506 | 0.17 | 1-Terpinen-4-ol; |
| 14 | 15.233 | 206371 | 0.11 | Thymol methyl ether |
| 15 | 16.255 | 152108 | 0.08 | Citronellyl formate |
| 16 | 17.154 | 980424 | 0.52 | 2-Methoxy-4-vinylphenol |
| 17 | 17.391 | 553057 | 0.29 | 2-Hydroxycineol; |
| 18 | 18.458 | 208787 | 0.11 | 1-Terpinenol; |
| 19 | 18.630 | 135193 | 0.07 | Neryl acetate |
| 20 | 18.706 | 214984 | 0.11 | Copaene; |
| 21 | 19.689 | 7150711 | 3.80 | Isocaryophyllene |
| 22 | 20.184 | 715315 | 0.38 | Isochavibetol |
| 23 | 20.449 | 1735258 | 0.92 | alpha-Humulene |
| 24 | 22.462 | 1564912 | 0.83 | 3',5'-Dimethoxyacetophenone |
| 25 | 23.588 | 937548 | 0.50 | Humulene epoxide |
| 26 | 24.005 | 761245 | 0.40 | Viridiflorol |
| 27 | 24.337 | 417371 | 0.22 | Andrographolide |
| 28 | 24.438 | 540854 | 0.29 | Dodecane |
| 29 | 24.706 | 660807 | 0.35 | Longifolene |
| 31 | 25.814 | 1247888 | 0.66 | Coniferol |
| 32 | 26.208 | 856113 | 0.45 | TETRADECANOIC ACID |
| 33 | 27.323 | 135392 | 0.07 | Squalene |
| 34 | 27.606 | 2421223 | 1.29 | Neophytadiene |
| 35 | 27.959 | 431955 | 0.23 | Pentadecanoic acid |
| 36 | 28.340 | 754011 | 0.40 | 3,7,11,15-Tetramethyl-2-hexadecen-1-ol |
| 37 | 29.086 | 683072 | 0.36 | Hexadecanoic acid, methyl ester |
| 38 | 29.295 | 356282 | 0.19 | Oleic Acid |
| 39 | 29.712 | 22470785 | 11.93 | n-Hexadecanoic acid |
| 40 | 30.111 | 3974845 | 2.11 | trans-Sinapyl alcohol |
| 41 | 30.899 | 337254 | 0.18 | 9-Eicosenoic acid, (Z)- |
| 42 | 31.252 | 1415971 | 0.75 | Palmitic acid |
| 43 | 31.759 | 753964 | 0.40 | Linoleic acid |
| 44 | 31.859 | 995581 | 0.53 | Linoleoyl Chloride |
| 45 | 32.026 | 3228940 | 1.71 | Phytol |
| 46 | 32.395 | 16550800 | 8.79 | Linoelaidic acid |
| 47 | 32.494 | 25543553 | 13.56 | 7-Tetradecenal, (Z)- |
| 48 | 32.823 | 4778228 | 2.54 | OCTADECANOIC ACID |
| 49 | 35.073 | 245968 | 0.13 | 1,7-Dioxaspiro[5.5]undec-2-ene |
| 50 | 37.795 | 2042438 | 1.08 | Palmitin |
| 51 | 40.295 | 727491 | 0.39 | 7-Tetradecenal, (Z)- |
| 53 | 40.829 | 725124 | 0.38 | 6-Methyl-3-pyridinamine |
| 55 | 41.709 | 419690 | 0.22 | Aristolediene |
| 56 | 42.755 | 551392 | 0.29 | Perhydrophenalene, (3a.alpha., 6a.alpha., 9a.alpha., 9b.bet |

| | | | | |
|---|---|---|---|---|
| 57 | 42.827 | 988138 | 0.52 | PHENOL, 4-[[4-[(3,4-DIMETHOXYPHENYL)METHYL |
| 58 | 44.394 | 393796 | 0.21 | 1-(3,4-Dimethoxyphenyl)decane-3,5-diyl diacetate |
| 59 | 45.967 | 526255 | 0.28 | 1-EICOSANOL |
| 60 | 46.361 | 4125811 | 2.19 | dl-.alpha.-Tocopherol |
| 61 | 48.430 | 6863539 | 3.64 | Campesterol |
| 62 | 49.054 | 6894571 | 3.66 | -Stigmasterol |
| 63 | 49.922 | 791443 | 0.42 | Cycloeucalenyl acetate |
| 64 | 50.653 | 17314378 | 9.19 | gamma.-Sitosterol |
| 65 | 51.271 | 4556673 | 2.42 | Lupeol |
| 66 | 51.703 | 9419116 | 5.00 | Cycloeucalenyl acetate |
| 67 | 52.682 | 1582302 | 0.84 | Lupeol |
| 68 | 52.906 | 501850 | 0.27 | Handianol |
| 69 | 53.080 | 613153 | 0.33 | 24-Norursa-3,12-diene |