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# GAS CHROMATOGRAPHY AND MASS SPECTROMETRY ANALYSIS OF *SOLANUM VILLOSUM* (MILL) (SOLANACEAE)

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**Keywords:**

GC –MS (Gas chromatography and Mass spectrometry), Anticancer, ethanol extract, *Solanum villosum*.

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**ABSTRACT:** In the present study the bioactive compounds of *Solanum villosum* leaves extract were used to evaluate by GC – MS method. 50g of fine powder was packed with No.1 Whatman filter paper and placed in soxhlet apparatus along with ethanol. The crude extract were collected and dried at room temperature, 30˚C after which yield was weighed and then performed to GC-MS analysis. The Ethanol leaf extracts of *Solanum villosum* to identified Twelve bioactive components such as, Methyl 11,14,17-Eicosatrienoate (29.49%), 4-(3,5-Di-Tert-Butyl-4-Hydroxy Phenyl) Butyl Acrylate (12.04%), N-Hexadecanoic acid (9.41%), Phytol (8.54%), 2H-1-Benzopyran-6-ol, 3,4-Dihydro-2,5,7,8-Tetra methyl-2-(4,8,12-Trimethyl Tridecyl)-Acetate,(2R)-( (5.70%), DL-Alpha Tocopherol (5.70%), Gamma – Tocopherol (3.55%), 3,4-Dihydro-3,5,8-Trimethyl-3-(4,8,12-Trimethyl Tridecyl)-(2H)1-Benzopyran-6-Acetate (3.55%), 3,7,11,15-Tetramethyl-2-Hexadecen-1-ol (3.38%), Cyclotrisiloxane, Hexamethyl (2.12%), Trimethyl(4-(1,1,3,3-Tetramethyl butyl)Phenoxy) silane (2.12%), Octasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11,13,13,15,15-Hexadecamethyl (1.97%) were present. In best of our knowledge, there is no documental evidence of gas chromatography and mass spectrum analysis to identify the chemical compounds from the plant species of *Solanum villosum.* This study helps to explore the potential compounds responsible for the biological activities of antimicrobial, antioxidant, antidiabetic, anticancer, anti-inflammatory, Diuretic and analgesic properties for application of drug formation in pharmaceutical fields.

**INTRODUCTION:** *Solanum* is one of the most important and largest genera of the family Solanaceae comprising of about 84 genera and 3000 species were identified throughout the Worldwide. *Solanum villosum* is also present in Western Ghats of Tamilnadu and varuious parts of India.

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The plants of *S. nigrum* complex has been traditionally used as an analgesic, antispasmodic, antiseptic, antidysentric, antinarcotic, emollient, diuretic, tonic, soporific, laxative, anticancer, antiulcer and for disorders of neuro-vegetative system etc.[1- 3] This medicinal value is mainly attributed to the alkaloidal contents of the plants.

*Solanum villosum* (Mill) belongs to family Solanaceae, it is commonly known as red-fruit nightshade, is widely distributed in many parts of India. The plant is an Ayurvedic herb with multiple medicinal properties. The plant *Solanum villosum*

contain many primary and secondary metabolites such as, alkaloids, flavonoids, phenols, saponins, tannins, terpenoids, steroids, carbohydrates, glycosides, amino acids and proteins [4]. The genus of *Solanum* species contains excellent antioxidant properties and free radical scavenging ability [5].

In recent years, gas chromatography and mass spectrography (GC–MS) has been applied unambiguously to identify the structures of different phytoconstituents from plant extracts and biological samples with great success. [6-7] Gas chromatography and mass spectrum is a reliable technique to identify the phytoconstituents of volatile matter, long-chain branched hydrocarbons, alcohols, acids and esters.[8] In this present study to analysis the bioactive compounds present in ethanol extract of *Solanum villosum* by Gas chromatography and mass spectrometry.

## MATERIALS AND METHODS:

**Plant material:** The leaves of the *Solanum villosum* (Mill) plant were collected from Thadagam hills at Coimbatore district, Tamilnadu, India. The specimen sample was authenticated by Dr.V.S.Ramachandran, Associate Professor, Department of Botany, Bharathiar University, Coimbatore, Tamilnadu, India. The voucher specimen was deposited in the herbarium center, Department of Botany, Bharathiar University, Coimbatore.

**Extraction of plant material:** Plant materials thoroughly washed and shade dried at room temperature after that grind into powder was packed with No.1 Whatman filter paper and placed in soxhlet apparatus along with ethanol. The crude extract were collected and dried at room temperature, 30˚C after which yield was weighed and then performed.

**GC-MS analysis:** Gas chromatography study includes the important optimization process such as i) introduction of sample extract onto the GC column, ii) separation of its components on an analytical column and iii) detection of target analysis by using mass spectrometric (MS) detector. 5 ml of ethanol extract was evaporated to dryness and reconstituted in to 2 ml methanol. The extracts were then subjected to GC-MS analysis. Chromatographic separation was carried out with instrument GC-MS-QP 2010 [SHIMADZU] instrument with Db 30.0 column (0.25µm diameter × 0.25um thickness). The oven temperature was programmed from 70 °C (isothermal for 5 min), with an increase of 10°C/min, to 200°C, then 5°C/min to 280°C, ending with a 35 min isothermal at 280°C. Mass spectra was taken at 70 eV; a scan interval of 0.5 s and Scan range from 40–1000 m/z. Helium was used as carrier gas at 99.999 % pressure with flow 1.0 ml/min and electronic pressure control on. Samples were dissolved in ethanol and injected automatically.

**Analytical condition:** Injection temperature at $240^0$C, interface temperature at $240^0$C and ion source temperature at $70^0$C were determined. Injection was performed in split less mode.

**Identification of compounds (Data analysis):** The mass spectra of compounds in samples were obtained by electron ionization (EI) at 70 eV and the detector operator in scan mode from 40 to 1000 m/z atomic mass units. Identification based on the Molecular weight, Molecular formula, Retention time and peak area %. It is done in order to determine whether this plant species contains any individual compound or group of compounds which may substantiate its current commercial and traditional use as herbal medicine, in addition to determine the most appropriate methods of extracting these compounds. These results will consequently be discussed in the light of their putative biological and therapeutic relevance.

## RESULTS AND DISCUSSION:

**Gas Chromatography-Mass Spectroscopy:** GC-MS is the best technique to identify the bioactive constituents of long chain hydrocarbons, alcohols, acids, ester, alkaloids, steroids, amino and nitrogen compound. The present research work deals with the ethanolic extracts of leaf extract of *Solanum villosum* to analysis Gas Chromatography-Mass Spectroscopy. The extracts are a complex mixture of many constituents totally twelve compounds identified (**Table 1** and **Fig 1).** In best of our knowledge and literature survey there is no report of gas chromatography and mass spectrum analysis to identify the chemical compounds from the plant species of *Solanum villosum.* GC-MS analysis showed the revealed that existence of the compounds such as, Methyl 11,14,17-

Eicosatrienoate (29.49%), 4-(3,5-Di-Tert-Butyl-4-Hydroxy Phenyl) Butyl Acrylate (12.04%), N-Hexadecanoic acid (9.41%), Phytol (8.54%), 2H-1-Benzopyran-6-ol, 3,4-Dihydro-2,5,7,8-Tetra methyl-2-(4,8,12-Trimethyl Tridecyl)-Acetate,(2R)-( (5.70%), DL-Alpha Tocopherol (5.70%), Gamma – Tocopherol (3.55%), 3,4-Dihydro-3, 5, 8 – Trimethyl – 3 - (4, 8, 12-Trimethyl Tridecyl) - (2H)1 – Benzopyran – 6 - Acetate (3.55%), 3, 7, 11, 15 – Tetramethyl – 2 - Hexadecen-1-ol (3.38%), Cyclotrisiloxane, Hexamethyl (2.12%), Trimethyl(4-(1,1,3,3-Tetramethyl butyl) Phenoxy) silane (2.12%), Octasiloxane,1,1,3,3,5,5,7,7,9,9,11,11,13,13,15,15-Hexadecamethyl (1.97%) were present. The compounds are identified with their retention time (RT), Molecular formula, Molecular weight, and concentration (peak area %) of the corresponding compounds. The GC-MS chromatogram of the seven peaks of the compounds was detected. Chromatogram GC-MS analysis of the ethanolic extract of *Solanum villosum* showed the presence of 3 major peaks and the components corresponding to the peaks were determined.

**TABLE 1: GAS CHROMATOGRAPHY AND MASS SPECTRUM ANALYSIS OF THE ETHANOL EXTRACT OF *SOLANUM VILLOSUM* (MILL)**

| S.No | Compound Name | % of Peak Area | Retention time (RT) | Molecular formula (MF) | Molecular weight (MW) | Nature of the compound | Biologial activity |
|---|---|---|---|---|---|---|---|
| 1. | 3,7,11,15-Tetramethyl-2-Hexadecen-1-ol | 3.386 | 16.83 | $C_{20}H_{40}O$ | 296 | Terpene alcohol | Cancer preventive, Antimicrobial, Antiinflammatory |
| 2. | N-Hexadecanoic acid | 9.416 | 18.300 | $C_{16}H_{32}O_2$ | 256 | Palmitic acid | Antioxidant, pesticide, flavour, 5-alpha reductase ihibitor,antifibrinolytic, Hemolytic, Lubricant, Nematicide, Antialopecic. |
| 3. | 4-(3,5-Di-Tert-Butyl-4-Hydroxy Phenyl) Butyl Acrylate | 12.04 | 18.43 | $C_{21}H_{32}0_3$ | 332 | - | Antiinflammatory |
| 4. | Phytol | 8.54 | 19.52 | $C_{20}H_{40}O$ | 296 | Phytol Diterpene alcohol | Antimicrobial, anticancer, antiinflammatory, Diuretic. |
| 5. | Methyl 11,14,17-Eicosatrienoate | 29.49 | 20.03 | $C_{21}H_{36}O_2$ | 320 | Unsaturated fatty acid ester | Antiarthritic, anticoronary, antiinflammatory. |
| 6. | Octasiloxane,1,1,3,3,5,5,7,7,9,9,11,11,13,13,15,15- Hexadecamethyl | 1.97 | 25.56 | $C_{16}H_{50}O_7Si_8$ | 578 | Volatile organic compounds | Anti microbial |
| 7. | 3, 4-Dihydro-3, 5, 8-Trimethyl-3-(4, 8, 12-Trimethyl Tridecyl)-(2H)1-Benzopyran-6-Acetate. | 3.55 | 26.86 | $C_{30}H_{50}O_3$ | 458 | Pentacyclic triterpenoids | Fungicides, insectiidess |
| 8. | Cyclotrisiloxane, Hexamethyl | 2.12 | 27.27 | $C_6H_{18}O_3Si_3$ | 222 | Phenolic compounds | Antimicrobial potential, Antimicrobial, antioxidants |
| 9. | 2H-1-Benzopyran-6-ol, 3,4-Dihydro-2,5,7,8-Tetra methyl-2-(4,8,12-Trimethyl Tridecyl)-Acetate, (2R) - ( | 5.70 | 27.49 | $C_{31}H_{52}O_3$ | 472 | Vitamin E acetate | antimicrobial anti oxidasnt |
| 10. | DL-Alpha Tocopherol | 5.70 | 27.49 | $C_{29}H_{50}O_2$ | 430 | Vitamin E | Preservative, flavor, antiseptic, stomach and internal hemorrhoids. |
| 11. | Trimethyl(4-(1,1,3,3-Tetramethyl butyl)Phenoxy) silane | 2.12 | 27.27 | $C_{17}H_{30}OSi$ | 278 | Methyl ether | Vitamin D, rickets and antioxidants |
| 12. | Gamma - Tocopherol | 3.55 | 26.86 | $C_{28}H_{48}O_2$ | 416 | Vitamin E | Analgesic, antidiabatic antiinflammatory, antioxidant, antidermatitic, antileukemic, antitumor, anticancer, hepatoprotective, anticoronary. |

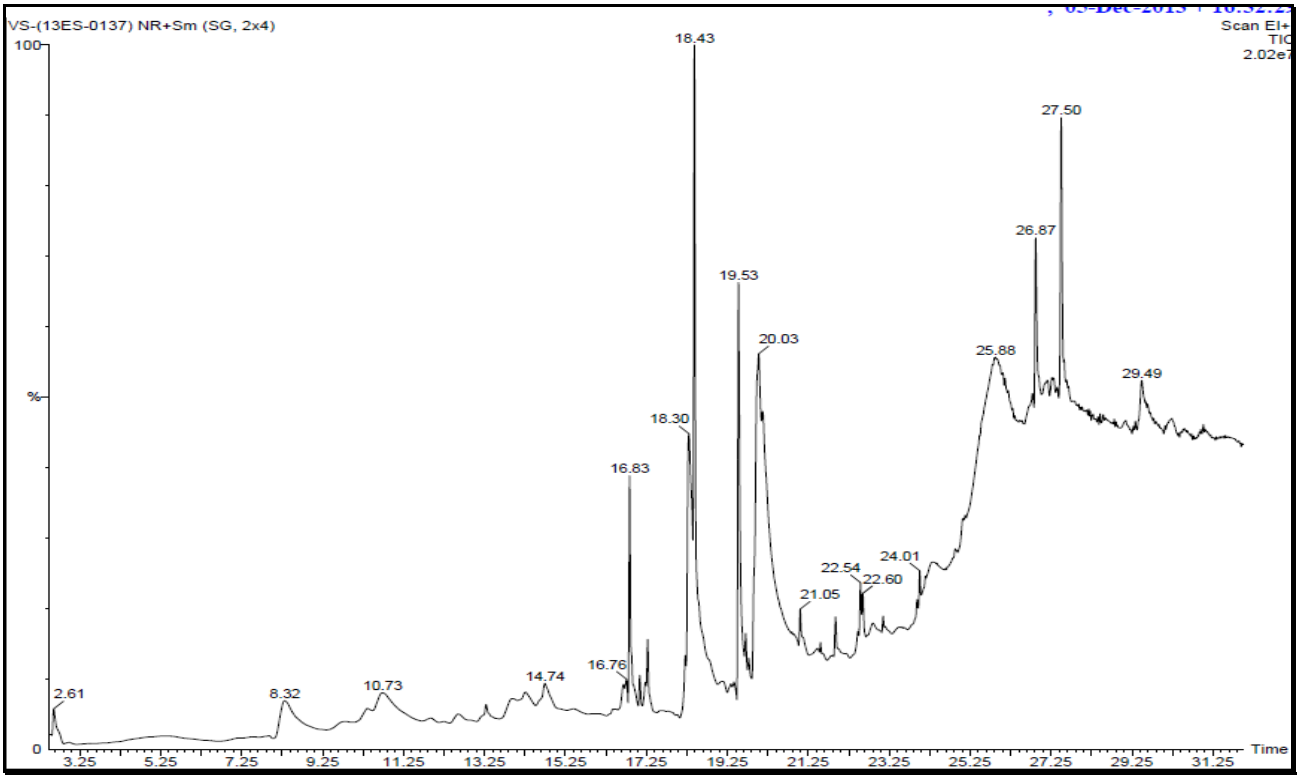


**FIG. 1: SHOWING GC-MS ANALYSIS OF ETHANOL EXTRACT OF *SOLANUM VILLOSUM* (MILL)**

Many medicinal plants are rich source of secondary metabolites such as alkaloids, phenol, glycosides, flavonoids, tannins and terpenoids determined by gas chromatography and mass spectrum [9-10]. The plant contain large amount of primary and secondary metabolites exert a wide range of biological activities on physiological systems. [11] It also reported that the activities of some plant constituents with compound nature of flavonoids, palmitic acid (hexadecanoic acid, ethyl ester and n-hexadecaonoic acid), unsaturated fatty acid and linolenic (docosatetraenoic acid and octadecatrienoic acid) as antimicrobial, anti-inflammatory, antioxidant, hypocholesterolemic, cancer preventive, hepatoprotective, antiarthritic, antihistimic, antieczemic and anticoronary [12].

Octadecanoic acid, Hexadecanoic acid and stigmasterol compounds have the property of antioxidant, antimicrobial, hypocholesterolemic, antiarthritic, anti-inflammatory [13.] Palmitic acid is reported to be an antioxidant, nematicide and a pesticide while Melitol and Phytol are said to be cancer preventive. The presence of phytol compounds attributes to the antimicrobial, anti-inflammatory and anticancer property of the plant leaves [14- 15]

The phenomenon of additive or synergistic effects is often crucial to bioactivity in plant extracts and in some cases; the activity is lost in purified fractions. Development of bacterial resistance to synergistic drug combinations, such as those found in plants, may be slower than for single drug therapies [16 -17]. It is believed that crude extracts from medicinal plants are more biologically active than isolated compounds due to their synergistic effects [18].

It was concluded that ethanol extract of *Solanum villosum* leaves possess various potent bioactive compounds and antidiabetic, analgesic, antiseptic, antidysentric, diuretic, antioxidant, anti-inflammatory, antiulcer and anticancer properties it is recommended as drug formation to pharmaceutical industries. Further studies are needed to explore the potential bioactive compounds responsible for the biological activities of *Solanum villosum*.

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