

ASSESSING THE BIOACTIVE CONSTITUENTS OF *DODONEA VISOSA* (VIRAALI) THROUGH GC-MS

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Abstract–The present effort was intended to investigate *Dodonea visosa* for phytochemical compounds and characterize the chemical constituents of plant using GC-MS. The shade dried leaf, stem, internode, callus parts of plant powder of *DodoneaVisosa* was extracted with Methanol overnight, filtered and concentrated. The GC Clarus 500 (Perkin Elmer) used in the investigation employed a column packed with Elite- 5MS (5%Diphenyl / 95% Dimethyl poly siloxane, 30mm x 0.25mm x0.25µmdf) and the components were separated using Helium (1ml/min) as the carrier gas. The 2 µl sample extract injected into the instrument was detected by the Turbo mass gold detector (Perkin Elmer) with the aid of the Turbomass 5.2 software. The GC-MS analysis provided different peaks determining the presence of seven phytochemicals for Viraali Leaf dry powder, two Phyto-chemicals for Viraali Stem dry powder, six Phytochemicals for Viraali Internode dry powder, three phytochemicals for Viraali Callus dry powder. The phytochemical and GC-MS profiling methanolic extract of *Dodonea visosa* revealed the presence of bioactive compounds with important medicinal properties. Hence, the presence of these phytochemicals could be responsible for the therapeutic effects of the plant.

INTRODUCTION

Plants are used as medicines in various cultures and serve as a source of many potent drugs due to the presence of certain bioactive compounds for pharmaceutical industries. Plants contain different phytochemicals, also known as secondary metabolites. (Olivia *et al.*, 2021). Phytochemicals are useful in the treatment of certain disorders by their individual, additive, or synergic actions to improve health (Zhang *et al.*, 2015).

Phytochemicals are vital in pharmaceutical industry for the development of new drugs and preparation of therapeutic agents (Ashraf, 2020). The development of new drugs starts with identification of active principles from the natural sources. The screening of plant extracts is a new approach to find therapeutically active compounds in various plant species (Atanasov *et al.*, 2021)

Phytochemicals such as flavonoids, tannins,

saponins, alkaloids, and terpenoids have several biological properties which include antioxidant, anti-inflammatory, anti-diarrhea, anti-ulcer, and anticancer activities, among others.

During the last decade, use of traditional medicine has prolonged globally and has gained attractiveness (Amudha and Rani, 2014). With the incredible expansion in the use of traditional medicine worldwide, safety and efficiency as well as quality control of herbal medicines and traditional therapies have become important concerns for both health authorities and the public (Ekor, 2014). There is still a significant lack of research data in this field. In the absence of pharmacopoeia data on the various plant extracts, it is not possible to isolate or standardize the active contents having the desired effects. Screening of active components from plants has direct relation to the development of new medicinal drugs which have efficient protection and treatment role against various diseases.

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Dodonea visosa is a shrub with various ethnomedical applications. The roots are used to treat wounds, leaves for gastrointestinal tract, skin and rheumatism, bone fracture, diarrhea, as antibacterial, antifungal, and as antidote for snakebite (Rajamanickam *et al.*, 2010). There is a report of its anti-HIV role. This plant is reported to have antiplasmodial, to cure sore throat and cold, as antidiabetic and anti-inflammatory. The plant is reported to have antioxidant and anticholinesterase activities (Mongalo *et al.*, 2020). The leaf extracts of this plant are reported to contain flavonoids, terpenoids, saponins, reducing sugars, and steroids.

Large number of medicinal plants and their purified constituents has shown beneficial therapeutic potentials. With this situation, this study was aimed to identify the phytoconstituents present in methanolic extract of *Dodonea visosa* using GC-MS analysis.

Gas chromatography-mass spectroscopy (GC-MS) is a combined analytical technique used to determine and identify compounds present in a plant sample. GC-MS plays an essential role in the phytochemical analysis and chemotaxonomic studies of medicinal plants containing biologically

active components.

MATERIALS AND METHODS

All the chemicals and reagents used for the research were of analytical grade. The leaf, stem, internode, callus parts of plant were collected from the natural habitats of Kannankudi village, Tevakottai Taluk, Sivangai District of Tamilnadu, India. The samples were washed thoroughly in running tap water to remove soil particles and adhered debris and finally washed with sterile distilled water. The leaf, stem, internode, callus parts of plant were shade dried and ground into fine powder. The powdered materials were stored in air tight polythene bags until use.

Fifty grams of powdered sample was extracted with methanol overnight and filtered through ash less filter paper with sodium sulphate and the extract was concentrated. The extract was analyzed using the Clarus 500 GC-MS (Perkin Elmer). 2 μ l of the methanolic extract of *Dodonea visosa* was employed for GC-MS analysis.

The Clarus 500 GC (Perkin Elmer) used in this analysis. It employed a fused silica column packed

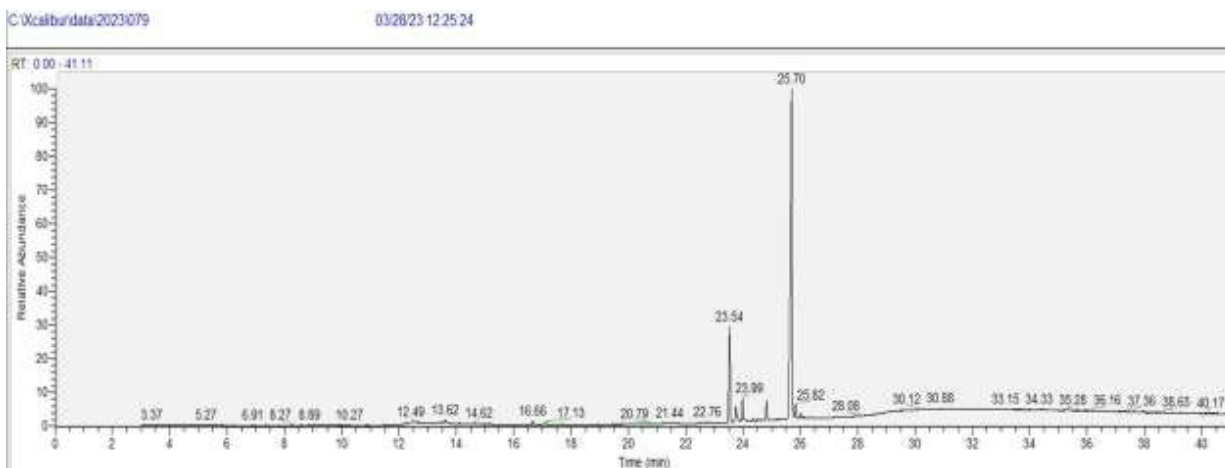


Fig 1. GC MS chromatogram of the methanolic extract of Viraali leaf dry powder

Table 1. Compounds identified in the methanolic extract of Viraali leaf dry powder

No	RT	Name of the compound	Molecular Formulae	Molecular Weight	Peak Area %
1	16.66	cis-5,8,11,14,17-Eicosapenta enoic acid	C ₂₀ H ₃₀ O ₂	302	0.47
2	23.54	(S,Z)-Heptadeca-1,9-dien-4,6 -diyn-3-ol	C ₁₇ H ₂₄ O	244	15.14
3	23.75	5,8,11,14-Eicosatetraenoic acid, methyl ester, (all-Z)-	C ₂₁ H ₃₄ O ₂	318	3.57
4	23.99 4	4,7,10-Hexadecatrienoic acid, methyl ester	C ₁₇ H ₂₈ O ₂	264	3.75
5	24.82	8,11-Octadecadiynoic acid, methyl ester	C ₁₉ H ₃₀ O ₂	290	2.91
6	25.69	10-Heptadecen-8-yenoic acid, methyl ester, (E)	C ₁₈ H ₃₀ O ₂	278	71.93
7	25.82	6,9,12-Octadecatrienoic acid, methyl ester	C ₁₉ H ₃₂ O ₂	292	2.23

with Elite -5MS (5%Diphenyl / 95% Dimethyl poly siloxane, 30mm x 0.25mm x0.25µm df) and the components were separated using helium as carrier gas at a constant flow of 1 ml/ min. The 2 µL sample extract injected into the instrument. It was detected by the Turbo gold mass detector (Perkin Elmer) with the aid of Turbo mass 5.2 software. During the GC process the oven was maintained at a temperature of 110 °C with 2 min holding. The injector temperature was set at 250°C. The different parameters involved in the operation of the Clarus 500 MS were also standardized. The Inlet line temperature was 200 °C. Mass spectra were taken at 70 eV; a scan interval of 0.5s and fragments from 45-450 Da. The MS detection was completed in 36 min. The detection employed the NIST ver. 2.0-year 2005 library.

RESULTS AND DISCUSSION

The results concerning to GC-MS analysis led to the identification of number of compounds from the GC fractions of the methanolic extract of *Dodonea visosa*. These compounds were identified through mass spectrum attached with GC. The active principles with their retention time (RT), molecular formula (MF), molecular weight (MW) and concentration (%) were tabulated in the given tables.

The results revealed that the presence of seven phytochemical for Viraali Leaf dry powder such as

cis-5,8,11,14,17-Eicosapenta enoic acid ,(S,Z)-Heptadeca-1,9-dien-4,6 -diyn-3-ol,5,8,11,14-Eicosatetraenoic acid methyl ester, (all-Z)-4,7,10-Hexadecatrienoic acid methyl ester, 8,11-Octadecadiynoic acid methyl ester, 10-Heptadecen-8-ynoic acid, methyl ester, 6,9,12-Octadecatrienoic acid, methyl ester (Table 1) (Fig 1).

Two phytochemicals for Viraali Stem dry powder such as (S,Z)-Heptadeca-1,9-dien-4,6 -diyn-3-ol, Alloaromadendrene oxide-(2). (Table 2) (Fig 2).

Six phytochemical for Viraali Internode dry powder such as 3-O-Methyl-d-glucose, D-glycero-D-manno-Heptitol, (S,Z)-Heptadeca-1,9-dien-4,6-diyn-3-ol, Androstan-17-one,3-ethyl-3-hydroxy-(5à), 5-Benzofuranacetic acid, 6-ethenyl-2,4,5,6,7,7a-hexah ydro-3,6-dimethyl-à-methylene-2-oxo-,methyl ester, Alloaromadendrene oxide-(2) (Table 3) (Figure 3).

Three phytochemicals for Viraali Callus dry powder such as 1,2-Benzenedicarboxylic acid, butyl 2- ethylhexyl ester, 1,8,9-Anthracenetriol, 3-methyl, 9,10-Anthracenedione, 1-hydroxy-4- methoxy (Table 4) (Figure 4).

Gas Chromatography- Mass Spectrometry (GC-MS) is a precious tool for reliable detection of bioactive constituents (Gomathi *et al.*, 2015). This study results were interpreted. By interpreting these compounds, it is found that *Dodonea visosa* possesses various therapeutical applications. The present

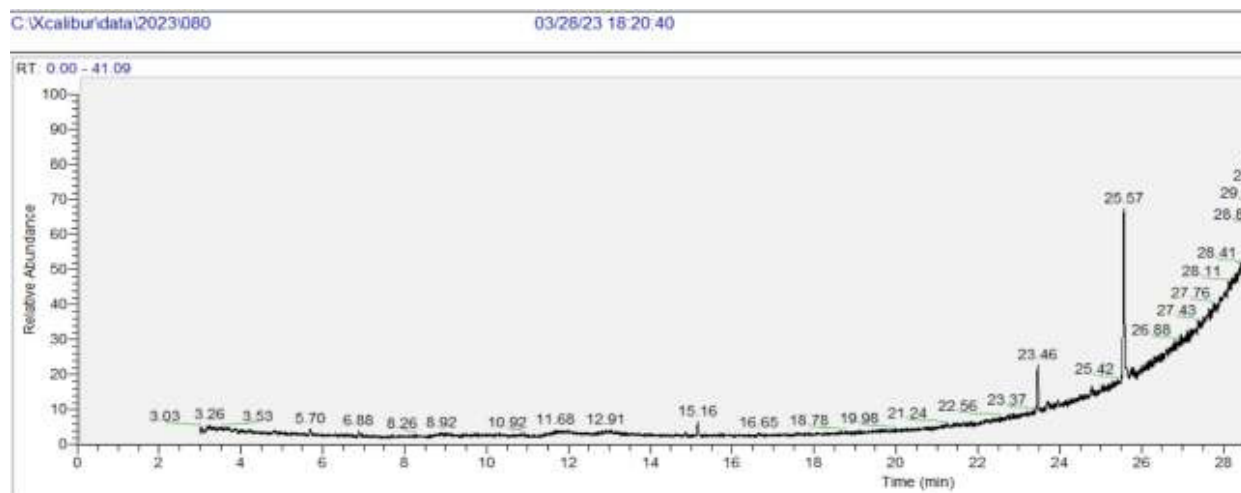


Fig. 2. GC MS chromatogram of methanolic extract of Viraali stem dry powder

Table 2. Compounds identified in the methanolic extract of Viraali stem dry powder:

No	RT	Nameofthecompoun	Molecular Formulae	Molecular Weight	Peak Area%
1.	23.46	(S,Z)-Heptadeca-1,9-dien-4,6-diyn-3-ol	C ₁₇ H ₂₄ O	244	21.15
2.	25.57	Alloaromadendreneoxide-(2)	C ₁₅ H ₂₄ O	220	78.85

study characterized the chemical profile of *Dodonea visosa* using GC-MS. The GC chromatogram shows the relative concentration of various compounds getting eluted as a function of retention time. The heights of the peak point out the relative concentration of the presented components. The mass spectrometer analyzes the compounds eluted at different times to identify the nature and structure of the compounds.

The investigation concluded that the stronger extraction capacity of methanol has produced number of active constituents responsible for many biological activities (Sobhyet *al.*, 2017). So, these might be utilized for the development of traditional medicines and further investigation needs to elute novel active compounds from the medicinal plants which may be created a new way to treat many incurable diseases including cancer (Ammal and Bai, 2013). To separate volatile substances in a mixture, gas chromatography is normally utilized (Chiu and Kuo, 2020).

Phytol have antioxidant, antiallergic

antinociceptive and anti-inflammatory activities. Recent studies have revealed that phytol is an excellent immunostimulant. It is superior to a number of commercial adjuvants in terms of longterm memory induction and activation of both innate and acquired immunity (Saha and Bandyopadhyay, 2020). Phytol showed antimicrobial activity against *Mycobacterium tuberculosis* and *Staphylococcus aureus*. Phytol was observed to have antibacterial activities against *Staphylococcus aureus* by causing damage to cell membranes as a result there is a leakage of potassium ions from bacterial cells. Phytol is a key acyclic diterpene alcohol that is a precursor for vitamins E and K1. It is used along with simple sugar or corn syrup as a hardener in candies.

Cyclohexene (5.09%): antioxidant and anti-diabetic. Methyl palmitate (18.56%) - used in textiles, spin, finishers, and food as an emulsifier or oiling agent. Detergents, soaps, shampoos, shaving creams, and other cosmetic goods are made with methyl stearate (12.57%) (Han *et al.*, 2023). Dibutyl

Table 3. Compounds identified in the methanolic extract of Viraali internode dry powder:

No	RT	Nameofthecompound	Molecular Formulae	Molecular Weight	Peak Area%
1.	12.23	3-O-Methyl-d-glucose	C ₇ H ₁₄ O ₆	194	1.74
2.	13.33	D-glycero-D-manno-Heptitol	C ₇ H ₁₆ O ₇	212	1.56
3.	23.50	(S,Z)-Heptadeca-1,9-dien-4,6-diyn-3-ol	C ₁₇ H ₂₄ O	244	20.19
4.	23.72	Androstan-17-one,3-ethyl-3-hydroxy-,(5à)-	C ₂₁ H ₃₄ O ₂	318	4.43
5.	23.96	5-Benzofuranaceti acid, 6-ethenyl-2,4,5,6,7,7a-hexah ydro-3,6-dimethyl-à-methylene-2-oxo-, methyl ester	C ₁₆ H ₂₀ O ₄	276	4.28
6.	25.64	Alloaromadendreneoxide-(2)	C ₁₅ H ₂₄ O	220	67.81

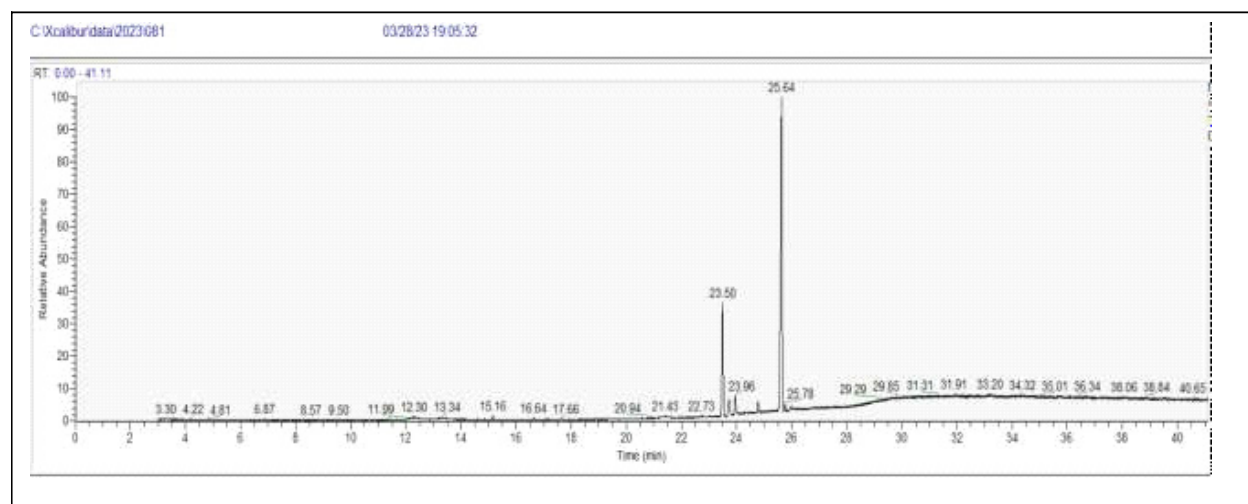
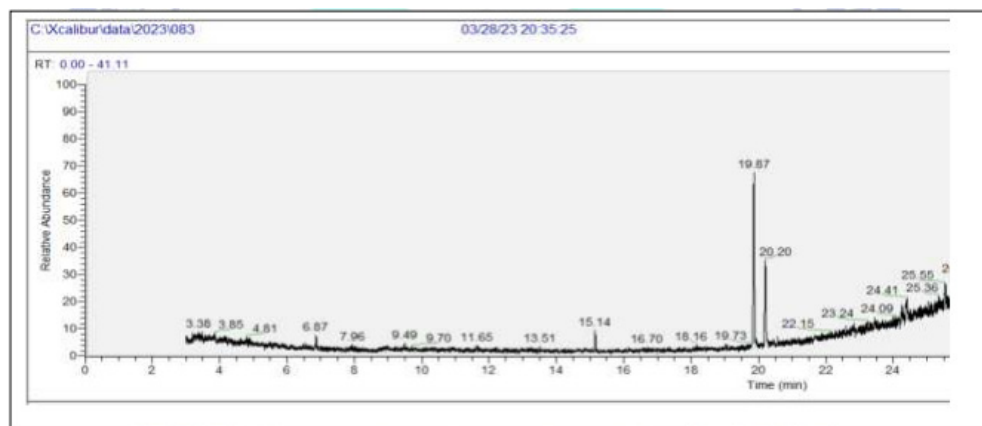


Fig 3. GC MS chromatogram of methanolic extract of Viraali internode dry powder

Table 4. Compounds identified in the methanolic extract of Viraali Callus dry powder:

No	RT	Name of the compound	Molecular Formulae	Molecular Weight	Peak Area %
1	15.14	1,2-Benzenedicarboxylic acid, butyl 2- ethylhexyleste	C ₂₀ H ₃₀ O ₄	334	8.98
2	19.87	1,8,9-Anthracenetriol, 3-methyl	C ₁₅ H ₁₂ O ₃	240	60.29
3	20.20	9,10-Anthracenedione, 1-hydroxy-4- methoxy	C ₁₅ H ₁₀ O ₄	254	30.73

**Fig. 4.** GC MS chromatogram of methanolic extract of Viraali Callus dry powder

phthalate (18.64%) was once used in nail polish, however, it is now prohibited. Elemicin (1.99 %) is a flavoring ingredient that can also be used medicinally in high dosages. Ketorolac (1.07 %) is an NSAID that is used to relieve acute pain for a brief period of time (5 to 7 days) following an injury, dental issue, surgery, or childbirth. These are the biochemical compounds which are used as an ingredient in sunscreens and cosmetics (Sathya *et al.* 2023). Octocrylene (1.42%) is also used as a component in hair products.

GC-MS-MS analysis identified 42 phytochemicals as components of *M. dubia* leaf extract. In the GC-MS-MS chromatogram, the peaks of the compounds were identified along with their retention time, molecular formula, molecular weight, and concentration (peak area percent) (Pangi *et al.*, 2023)

The compounds discovered in the previous study were octadecanoic acid (15.71%), hexadecanoic acid (11.10%), humelene (3.24%), caryophyllene (6.07%), aromadendrene (3.53%), and germacrene-D (2.89%), which has been shown to have pesticidal activity by (Murugesan *et al.*, 2013).

The compounds discovered in the current study were octadecanoic acid (15.71%), hexadecanoic acid (11.10%), humelene (3.24%), caryophyllene (6.07%), aromadendrene (3.53%), and germacrene-D (2.89%), which has been known to have pesticidal activity.

The main components of the essential oil extracted from *Vitex pseudo-negundo* leaves were 2,6,6-Trimethylbicyclo [3.1.1] hept-2-ene (18.2 %), 1,8-Cineole (16.7 %), Phenol, bis(1,1-dimethylethyl), Viridiflorol (8.32 %), dl-Limonene (4.4%) (1.31 %) (Ahmadvand *et al.*, 2014)

The compounds 9,12,15- Octadecatrienoic Acid, Methyl Ester reported that Antiinflammatory, Hypocholesterolemic, Cancer preventive, Hepato protective, Nematicide, Insectifuge, Antihistaminic, Antiarthritic, Anticoronary, Antieczemic Antiacne, 5-Alpha reductase inhibitor Antiandrogenic. Phytol Isomer is reported that it can be used for Anticancer, Anti-inflammatory Hypocholesterolemic, Nematicide and Anticoronary.

CONCLUSION

Several phytochemical evaluations have been carried out in different parts of *DodoneaVisosa* using GC-MS. This analysis showed the existence of various compounds with different chemical structures. The occurrence of various bioactive compounds proves the purpose of *Dodonea visosa* for various disorders. However, selection of individual phytochemical constituents may proceed to find an innovative drug. Hence, this type of effort will be supportive for in depth study.

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