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GC-MS Analysis of Phytochemical Compounds Present in the Methanolic Extract of *Viburnum punctatum* Buch-Ham Ex D.Don.

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ABSTRACT

Viburnum Punctatum aerial parts extract was prepared in methanolic solvent to study the phytochemical profile using Gas Chromatography-Mass Spectrometry method. GC-MS analysis of *Viburnum Punctatum* aerial parts extract revealed the existence of the major peaks presented in methanol were dl-Threitol (RT: 5.875 & Area%: 6.53), 4-Acetyl butyric acid (RT: 14.79 & Area%: 55.07), n-Hexadecanoic acid (RT: 17.58 & Area%: 3.21), Inositol (RT: 18.62 & Area%: 3.62), 9,12,15-Octadecatrienoic acid (Z,Z,Z)- (RT: 19.25 & Area%: 8.63), Benzyl β -d-glucoside (RT: 20.32 & Area%: 2.14) and Salicin (RT: 22.92 & Area%: 6.68). From this study it is obvious that *Viburnum Punctatum* aerial parts extract contains many biologically active compounds and also it gives a detailed insight about the phytochemical profile which could be exploited for the development of plant based drugs.

Keywords: *Viburnum punctatum*, GC-MS, Phytochemical Compounds.

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INTRODUCTION

A diverse range of bioactive molecules are produced by the plant which makes them an enriched source of different varieties of medicines. The ancient scholastic works included in the Atharva veda, Charaka and Sushruta comprises a rich heritage of knowledge to preventive and curative medicines. Natural products play an important role in drug development programs in the pharmaceutical industry [1] and almost 50% of all modern drugs are of natural origin [2]. Due to the efficacy and cost effectiveness of herbal drugs, it gained much importance in the recent years.

The present study on the medicinal plant namely *Viburnum punctatum* belongs to the family Caprifoliaceae and this plant is small trees, monotypic genus *Viburnum*, native to India, Indonesia, Bhutan, Cambodia, Nepal, Thailand, Vietnam and China. Asian *Viburnum* features dainty lymes of creamy white flower at the ends of the branches from early to mid-spring. It has dark green foliage throughout screen. The red fruits are held in abundance in spectacular clusters in mid-summer, expected to live for 40 years or more [3, 4]. The leaves were traditionally used for the treatment for fever, stomach disorder and mentioned to possess antiperiodic effect. The preliminary phytochemical investigation shows presence of flavonoids, alkaloids, glycosides, phenolic compounds, phytosterols and saponins [5, 6].



Figure 1: The Plant *Viburnum Punctatum*

In the last few years, Gas chromatography-mass spectrometry (GCMS) has become firmly established as a key technological platform for secondary metabolite profiling in both plant and non-plant species [7, 8, 9].

Gas chromatography-mass spectrometry (GC-MS) is a method that combines the features of gas-liquid chromatography and mass spectrometry to identify the different substances within a test sample. However, few reports are available with respect to the pharmacological properties of the plant [10]. Keeping this in view, the present study has been undertaken to identify the phytochemical compounds present in methanolic extract of *Viburnum punctatum* using GC-MS analysis.

MATERIALS AND METHODS

Plant Material

Aerial parts of *Viburnum punctatum* were collected from Kalakkad-Mundenthurai, Thirunelveli in the month of June 2013. The plant was authenticated by Prof. V. Chellathurai, Former Professor, Govt. Sidha Medical College, Thirunelveli. A voucher specimen of *Viburnum punctatum* was registered number is VPC-III /09/2013/14. The plant material was dried at room temperature, pulverized by a mechanical grinder, sieved through 40 mesh and stored in an air tight and light resistant container for further use.

Preparation of Extract

The coarsely powdered plant material was first defatted with Petroleum ether using Soxhlet apparatus. The extract was concentrated using rotary evaporator to get solid residue. The marc from the

central compartment was removed, dried and successively extracted with a series of solvents of increasing polarity with soxhlet extractor was done. Solvents used with increasing polarity are Chloroform, Methanol and Water [11, 12].

GC-MS Analysis

The phytochemical investigation of methanol extract of *Viburnum punctatum* was performed on a GC-MS-5975C (AGILENT) instrument.

GC CONDITION	
Column oven Temperature	70°C
Injector Temperature	240°C
Injector Mode	Split
Split Ratio	10
Flow Control Mode	Linear Velocity
Column Flow	1.51 ml/min
Carrier Gas	Helium 99.9995% purity
Injection Volume	1 microlitre

MS CONDITION	
Ion Source temp	200°C
Interface temp	240°C
Scan Range	40-1000m/z
Solvent cut time	5 mins
MS Start time	5 (min)
MS End time	35 (min)
Ionization	EI (-70ev)
Scan speed	2000

Identification of components

Total GC running time was 35 min and the interpretation on mass spectrum of GC-MS was done using the database of National Institute Standard and technology (NIST). The mass spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST. The name, molecular weight and structure of the components of the test materials were ascertained [13, 14].

RESULTS AND DISCUSSION

GC-MS chromatogram of the methanolic extract of *Viburnum punctatum* showed 21 peaks indicating the presence of 21 compounds. The chemical compounds identified in the methanolic extract of the aerial parts of *Viburnum punctatum* presented in table 1. GC-MS analysis revealed that the presence of 2,4-dihydroxy-2,5-dimethyl-3(2H)-furan-3-one, erythritol, 1,3,5-Triazine-2,4,6-triamine, 4-H-pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl, benzofuran 2,3-dihydro, octa-β-D-glucopyranoside, ethyl-∞-d-glucopyranoside, 1,1-biphenyl 4-chloromethyl, octadecanoic acid, Bis(2-ethylhexyl) phthalate and Arsenous acid are showed as minimum percent. dl-threitol, Salicylic acid, 1-deoxy-d-arabitol, n-hexadecanoic acid, inositol, 9,12,15-octadecatrienoic acid, benzyl-β-d-glucoside, and salicin are recorded predominantly. 4-acetyl butyric acid is maximum percent. Carbohydrates like D-flucose and lactose are considered amount is present. The GC-MS analysis revealed that the methanolic extract is mainly composed of oxygenated hydrocarbons, phenolic hydrocarbons and nitrogen containing compounds. These phytochemicals are responsible for various pharmacological actions like antimicrobial and antioxidant activities.

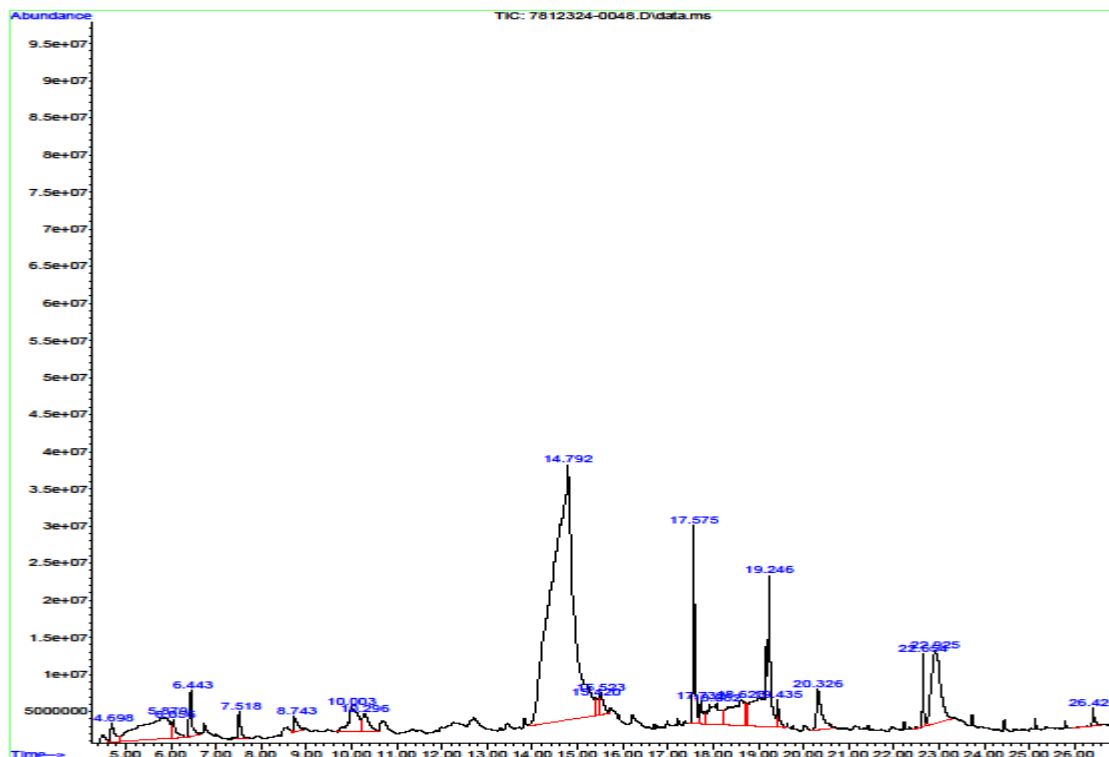
Viburnum punctatum is a potential folklore medicinal plant used for many diseases and infections. Phytochemical analysis by GC-MS revealed the presence of fatty acid esters, fatty acid amide, phenolic compounds, oxygenated hydrocarbons and nitrogen containing compounds as major compounds in methanolic fraction. Compositional variation in quantities, qualities and structural features may influence compounds behaviour on GC-MS as well as bioactivities of their precursor fractions.

Table 1: Phytocomponents identified in the methanol extract of *Vburnum punctatum* by GC-MS Peak report.

RT	Compound Name	Molecular Formula	Molecular Weight	Area %
4.69	2,4-dihydroxy-2,5-dimethyl-3(2H)-furan-3-one	C ₆ H ₈ O ₄	144.12	0.76
5.87	dl-Threitol	C ₄ H ₁₀ O ₄	122.12	6.53
6.05	Erythritol	C ₄ H ₁₀ O ₄	122.12	0.79
6.44	1,3,5-triazine-2,4,6-triamine	C ₃ H ₆ N ₆	126.11	1.30
7.51	4-H-pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl	C ₆ H ₈ O ₄	144.12	0.70
8.73	Benzofuran 2,3-dihydro	C ₈ H ₇ O	119.14	0.57
10.00	Salicylic acid	C ₇ H ₆ O ₃	138.12	2.35
10.29	1-Deoxy-d-arabitol	C ₅ H ₁₂ O	136.15	1.24
14.79	4-Acetyl butyric acid	C ₆ H ₁₀ O ₃	130.14	55.07
15.41	Octa-β-D-glucopyranoside	C ₁₄ H ₂₈ O ₆	292.36	0.74
15.51	Ethyl-∞-d-glucopyranoside	C ₈ H ₁₆ O ₆	208.20	0.78
17.58	n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256.42	3.21
17.72	1,1-biphenyl 4-chloromethyl	C ₁₄ H ₁₂ Cl ₂	251.15	0.82
18.08	Myo-Inositol	C ₆ H ₁₂ O ₆	180.15	2.26
18.62	Inositol	C ₆ H ₁₂ O ₆	180.15	3.62
19.25	9,12,15-Octadecatrienoic acid (Z,Z,Z)	C ₁₈ H ₃₀ O ₂	278.45	8.63
19.44	Octadecanoic acid	C ₁₈ H ₃₆ O ₂	284.47	0.64
20.32	Benzyl-β-d-glucoside	C ₁₃ H ₁₈ O ₆	270.27	2.14
22.64	Bis-2-ethyl hexyl phthalate	C ₂₄ H ₃₈ O ₄	390.55	0.78
22.92	Salicin	C ₁₃ H ₁₈ O ₇	286.27	6.68
26.42	Arsenous acid,tris ester	C ₃ H ₉ AsO ₃	168.02	0.38

Figure 2: GC-MS Profile of methanol extract of *Viburnum punctatum*

File :D:\NOV-12\20-11-12\7812324-0048.D
 Operator : ESVARAIYA
 Acquired : 20 Nov 2012 19:40 using AcqMethod UNKNOWN METHOD NEW.M
 Instrument : GCMS
 Sample Name: 1:1 SAMPLE
 Misc Info :
 Vial Number: 3





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