GC-MS DETERMINATION OF BIOACTIVE CONSTITUENTS OF *PHOEBE WIGHTII* MEISN. (LEAF)

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Abstract

Plants are a rich source of secondary metabolites with interesting biological activities. In general, these secondary metabolites are an important source with a variety of structural arrangements and properties. The aim of this study is to determine the organic compounds present in the active fraction of *Phoebe wightii* plant extract with the aid of GC-MS technique, which may provide an insight in its use in traditional medicine. The selected plant P. wightii is an endemic species which belongs to family Lauraceae. The investigation was carried out to determine the chemical components of *P. wightii* using Perkin-Elmer Gas Chromatography–Mass Spectrometry, while the mass spectra of the compounds found in the extract was matched with the National Institute of Standards and Technology (NIST) library. GC/MS analysis of ethanol extract of *P. wightii* leaves revealed the existence of Ethoxycarbonyl, 2-bromobenzoyl,Triisopropylsilyl, 4-chlorophenyl,1-Eicosene, Octadecanoic acid, ethyl ester, hexadecanoic acid, acetyloxy, O- Methyloxime,4-chlorobenzoyl,4-chlorophenyl,hydroxymethyl, hexadecane etc. The results of this study offer a platform of using *P. wightii* as herbal alternative for various diseases. The plant leaf of *P.wightii* screened for bioactive compounds seemed to have the potential to act as a source of useful drug and also to improve the health status of the as a result of the presence of a various compounds that are vital for good health. It also holds for the production of novel drugs with isolation of specific compounds.

Keywords: consumers, isolation, *Phoebe wightii* and secondary metabolites.

INTRODUCTION

Plants are a rich source of secondary metabolites with interesting biological activities. In general, these secondary metabolites are an important source with a variety of structural arrangements and properties (Kensa and Neelamegam, 2016). Ayurveda stresses the use of plant-based medicines and treatments. But when compared the Chinese medicine is more established than Ayuvedic medicine. This is due to even after Chinese people migrating to other countries they still follow their own culture. And also the Chinese people wherever in the world are actively participating in export and import of their medical system (Aneesh *et al.*, 2009). It is a sad fact that nowadays we are moving away from nature and due to our

undisciplined life style new diseases are being identified. But the fact is that our rich nature contains remedy for all diseases. Potentially valuable treasures in medicinal plants remain unexplored. By considering the scope of these medicinal plants we have to use more amounts of time and resources into developing medicines by medicinal plants. If we can come back to our nature, culture and tradition on use of medicinal plants it can bring up a bright and healthy new generation (Kirtikarand Basu, 1918).

Phoebe wightii is a tree commonly found in wasteland of garden and plains. It is a monotypic to genus, native to mexico. It belongs to the family Lauraceae. The investigation was carried out to determine the chemical components of *Phoebe wightii* leaf using Perkin Elmer Gas chromatography-Mass spectrometry. Higher plants as source of bioactive compounds continue to play a dominant role in the maintenance of human health. Reports available on green plants represent a reservoir of effective chemotherapeutants these are non-phytotoxic, more systematic and easily biodegradable (Vyas., 1999; Kaushik *et al*., 2002; ChamanLal and Verma., 2006).

Gas Chromatography Mass Spectroscopy, a hyphenated system which is a very compatible technique and the most commonly used technique for the identification and quantification purpose. The unknown organic compounds in a complex mixture can be determined by interpretation and also by matching the spectra with reference spectra (Ronald Hites, 1997)

In the last few years, gas chromatography mass spectrometry (GC-MS) has become firmly established as a key technological platform for secondary metabolite profiling in both plant and non-plant species (Robertson, 1991; Fernie *et al*., 2004; Kell *et al*., 2005). Gas chromatography – mass spectrometry (GC-MS) is a method that combines the features of gas-liquid chromatography and mass spectrometry to identify different substances within a test sample. However, few reports are available with respect to the pharmacological properties of the plant. Keeping this in view, the present study has been undertaken to investigate the antibacterial effects and identify the phytoconstituents present in ethanolic leaf extracts of *Phoebe wightii* using GC-MS analysis.

MATERIALS AND METHODS

METHODOLOGY

Collection of plant sample

Leaf of *P. wightii* was collected from Kotagiri, Coimbatore of Tamil Nadu, India and authenticated by Botanist Dr. R. Murugan, BSI, Southern circle, Kovai, India. A voucher specimen was deposited in the herbarium of the Botanical Survey of India Coimbatore; Herbarium code No. BSI/SRC/19/710-20/Tech.

Plant sample extraction

Leaves were cleaned, shade dried and pulverized to powder in a mechanical grinder. Required quantity of powder was weighted and transferred to stoppered flask and treated with

ethanol until the powder is fully immersed. Dark green residues were obtained after concentrating the extract under reduced pressure. The obtained extracts were stored in desiccators for further GC-MS.

GC-MS ANALYSIS

GC-MS analysis was carried out on a GC clarus 500 Perkin Elmer system comprising a AOC-20i auto sampler and gas chromatograph interfaced to a mass spectrometer instrument employing the following conditions: column Elite-1 fused silica capillary column (30 x 0.25 mm ID x 1 µMdf, composed of 100% Dimethylpolysiloxane), operating in electron impact mode at 70 eV; Helium gas (99.9%) was used as carrier gas at a constant flow of 1 ml /min and an injection volume of 0.5 µI was employed (split ratio of 10:1) injector temperature 250°C; ion-source temperature 280°C. The oven temperature was programmed from 110°C (isothermal for 2 min), with an increase of 10°C/min, to 200°C, then 5°C/min to 280°C, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70 eV; a scan interval of 0.5 seconds and fragments from 40 to 450 Da. Total GC running time is 36 min. min. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. Software adopted to handle mass spectra and chromatograms was a TurboMassVer 5.2.0.

Identification of bioactive components

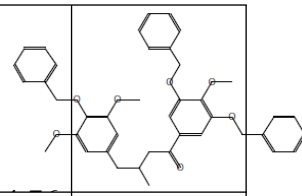
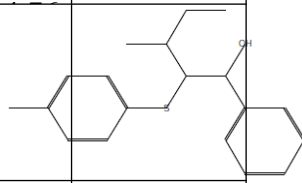
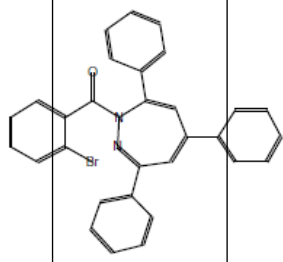
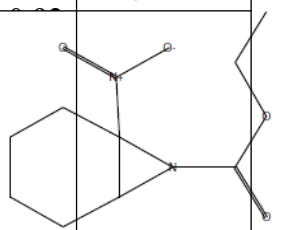
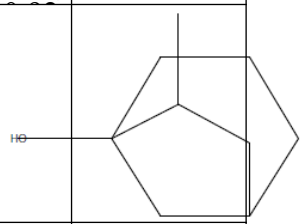
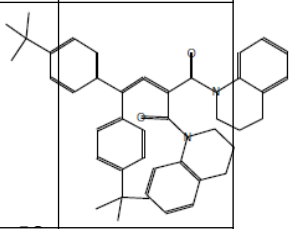
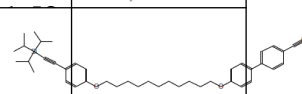
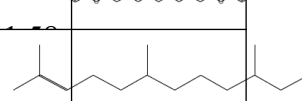
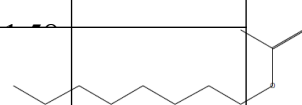
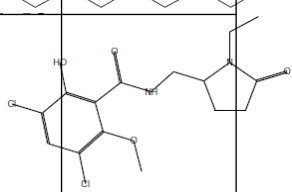
The relative percentage amount of each component was calculated by comparing its average peak area to the total peak areas. The detection employed the NIST (National Institute of Standards and Technology) Ver. 2.53 – year 2005 library. The compound prediction is based on Dr. Duke's phytochemical and Ethno botanical Database (Dukes, 1955) by Dr. Jim Duke of the Agricultural Research Service. Interpretation of GC-MS was conducted using the database of NIST having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight, and structure of the components of the test materials were ascertained.

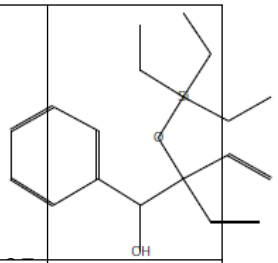
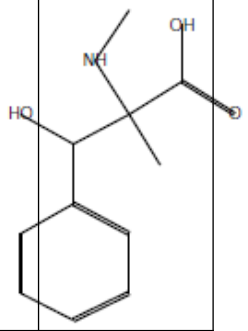
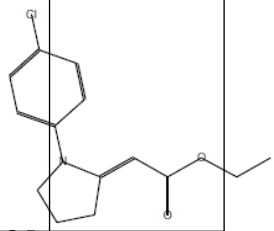
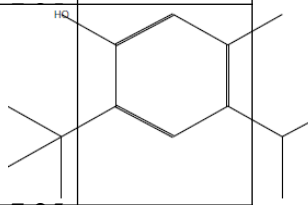
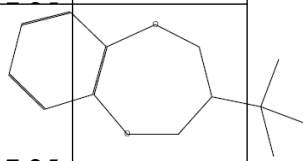
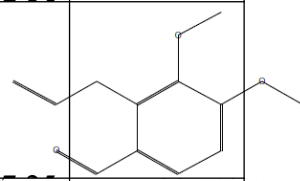
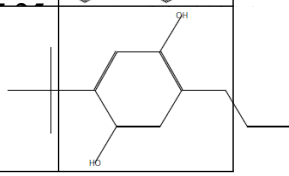
Results and Discussion

The GC-MS chromatogram of ethanolic extracts of leaf of *Phoebe wightii* revealed the presence of various compounds with corresponding peaks at different retention time GC-MS is one of the best techniques to identify the constituents in plants. The GC-MS analysis of *P. Wightii*leaf revealed the presence of 17 compounds. Table 1and figure 1 showed the various bioactive compounds were characterized and identified.

Table 1. GC-MS analysis of phytocomponents and their activities in the leaf of ethanolic extracts of *PHOEBE WIGHTII*.

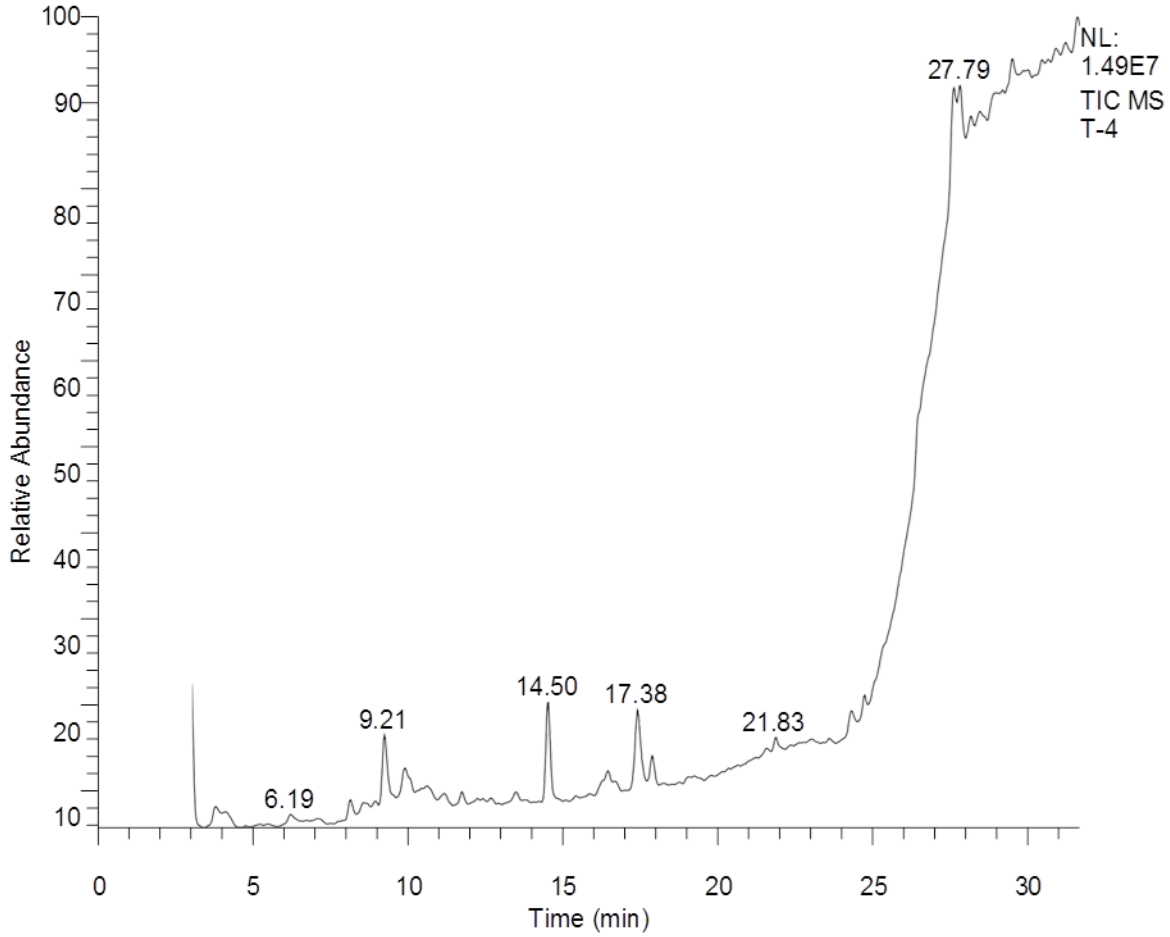
Sl. No.	Compound Name	Probability	Molecular Formula	Molecular Weight	Area %	Structure
1	4-(4-Benzyloxy-3,5-	18.48	C41H42O7	646	4.76	

	dimethoxyphenyl) 1-(3,5-dibenzy loxy-4-methoxyphenyl)-3-methylbutan-1-one					
2	(+)-(1r,2s,3r)-3-methyl-1-phenyl-2-(p-tolylthio)-1-pentanol	4.85	C19H24OS	300		
3	1-(2-Bromobenzoyl)-3,5,7-triphenyl-1H-1,2-diazepine	57.23	C30H21BrN2O	504		
4	7-(Ethoxycarbonyl)-8-nitroabicyclo[4.1.0]octane	2.41	C9H14N2O4	214		
5	7-methylbicyclo[3.2.1]octane-1-ol	2.03	C9H16O	140		
6	1,1-bis(4'-t-Butylphenyl)-3,3-bis(1',2',3',4'-tetrahydro-1'-quinolinyl carbonyl)-1,2-propadiene	31.03	C43H46N2O2	622		
7	4-{4-[(11-{4-[2-(Triisopropylsilyl)ethynyl]phenoxy}undecyl)oxy]phenyl}benzonitrile	6.25	C41H55NO2Si	621		
8	(6S,10S)-2,6,10-Trimethyl-2-dodecene	3.03	C15H30	210		
9	1-octylethanoate	2.44	C10H20O2	172		
10	(S)-5-[(3',5'-Dichloro-2'-hydroxy-6'-methoxybenzamide)methyl]-1-ethyl-2-pyrrolidone	1.92	C15H18Cl2N2O4	360		
11	1-Phenyl-2-triethylsilyloxy-2-vinyl-3-buten-1-ol	4.77	C18H28O2Si	304	0.97	

					
12	(2R,3R)-2,N-Dimethyl-3-phenylserine	3.75	C11H15NO3	209	
13	1-(4-Chlorophenyl)-2-[(ethoxycarbonyl)methylene]pyrrolidine	2.94	C14H16ClNO2	265	
14	2-tert-Butyl-4-isopropyl-5-methylphenol	11.88	C14H22O	206	
15	3,4-Dihydro-2H-1,5-(3"-t-butyl)benzodioxepine	9.58	C13H18O2	206	
16	2-Allyl-3,4-dimethoxybenzaldehyde	9.20	C12H14O3	206	
17	2-Allyl-5-t-butylhydroquinone	8.85	C13H18O2	206	

Source: Dr. Duke's phytochemical and ethnobotanical databases.

Fig. 1. GC-MS chromatogram of the ethanol extract of *PHOEBE WIGHTII*.



The identification of the phytochemical compounds was confirmed based on the peak area, retention time. The results revealed the4-(4-Benzyloxy-3,5-dimethoxyphenyl) 1-(3,5-dibenzy loxy-4-methoxyphenyl) -3-methylbutan-1-one,(+)-(1r,2s,3r)-3-methyl-1-phenyl-2-(p-tolylthio)-1-pentanol,1-(2-Bromobenzoyl)-3,5,7-triphenyl-1H-1,2-diazepine,7-(Ethoxycarbonyl)-8-nitroabicyclo[4.1.0]octane,7-methylbicyclo[3.2.1]octane-1-ol,1,1-bis(4'-t-Butylphenyl)-3,3-bis(1',2',3',4'-tetrahydro-1'-quinolinyl carbonyl)-1,2-propadiene,4-{4-[(11-{4-[2-(Triisopropylsilyl)ethynyl]phenoxy}undecyl)oxy]phenyl}benzonitrile,(6S,10S)-2,6,10-Trimethyl-2-dodecene 1-octylethanoate (S)-5-[(3',5'-Dichloro-2'-hydroxy-6'-methoxybenzamide)methyl]-1-ethyl-2-pyrrolidone,1-Phenyl-2-triethylsilyloxy-2-vinyl-3-buten-1-ol,(2R,3R)-2,N-Dimethyl-3-phenylserine,1-(4-Chlorophenyl)-2-[(ethoxycarbonyl)methylene]pyrrolidine,2-tert-Butyl-4-isopropyl-5-methylphenol,3,4-Dihydro-2H-1,5-(3"-t-butyl)benzodioxepine,2-Allyl-3,4-dimethoxybenzaldehyde,2-Allyl-5-t-butylhydroquinonerevealed the presence of various compounds with corresponding peaks at different retention time. The GC-MS analysis of *Phoebe wightii* leaf revealed the presence of 17 compounds (phytochemical constituents).

Plants synthesize an extensive array of secondary metabolites often highly compound structures. The chemical investigations of medicinal plants have largely been driven to find

new drugs to treat human disease. The secondary metabolites have been of interest to humans as flavors, fragrance, dyes, pesticides and pharmaceuticals (Govindaraj and Rajangam, 2017).

Conclusion

In this present study about 17 bioactive compounds are identified from ethanol extract of *P. wightii* by GC-MS method. The presence of various phytoactive compounds in this plant is responsible for the pharmaceutical properties. Therefore, it is recommended as a plant of phytopharamaceutical importance. Present study may be useful in the identification of novel drugs from stem of *P. wightii*. It is concluded that the ethanol can be used for extracting active compounds from plants and incorporating into medicinal food products. In addition further research is necessary to identify the active compounds responsible for therapeutic activity and animal study to evaluate the dosage of the identified chemical compounds.

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