



GC-MS ANALYSIS OF BIOACTIVE COMPOUNDS PRESENT IN ETHANOLIC STEM BARK EXTRACT OF AN ENDEMIC PLANT *CHIONANTHUS MALA-ELENGI* (DENNST.) P.S. GREEN (OLEACEAE)

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Abstract

Traditional and herbal medicines are the most easily available health care remedies. *Chionanthus mala-elengi* (Dennst.) P. S. Green is an endemic tree species of the family Oleaceae. It is used in giddiness, epilepsy, wound healing and liver disease. Phytochemical studies of this plant have not been reported so far. The aim of the present study is to characterize the chemical constituents present in ethanolic stem bark extract of *Chionanthus mala-elengi*. Thirty grams of powdered stem bark was extracted using 150 ml ethanol in Soxhlet apparatus. The mixture was filtered. The filtrates were evaporated and the dried ethanolic stem bark extract was used for further studies. Gas Chromatography - Mass Spectrometry analysis provided different peaks indicating the presence of sixteen different phytochemical compounds. Thus, the qualitative determination of stem bark ethanolic extract of *C. mala-elengi* using GC-MS analysis revealed the presence of various bioactive compounds which is used for different ailments by traditional practitioners.

Key words : *Chionanthus mala-elengi*, stem bark, GC-MS, bioactive compounds.

Introduction

Medicinal plants are used to cure various diseases throughout the world. They possess various ranges of chemical constituents with pharmacological and biological activities. They have around 1000 bioactive principles and mainly contain alkaloids, tannins, flavanoids and phenolic compounds (Al-Ansari *et al.*, 2019). These secondary metabolites are important components of pharmaceutical industry for drug development and therapeutic agents (Omorowa *et al.*, 2015). Plant kingdom is an important source which produces a wide range of natural antioxidants (Hossain and Shah, 2015).

Screening of active compounds from the plants had lead to the invention of new medicinal drugs (Omorowa *et al.*, 2015). Recently many scientists and researchers have focused their attention to isolate the active pure compound from the plant extracts which have been used in herbal and traditional remedies (Hossain *et al.*, 2013). Extraction is used to separate the medicinally active portion of plant tissue using selective solvents. It is the main step for isolating the individual chemical entities (Handa *et*

al., 2008). GC-MS analytical technique is applied in various fields such as food, beverage, flavor, fragrance analysis, medicine and pharmaceutical applications (Chauhan *et al.*, 2014). Gas chromatography is used to separate the compounds individually, whereas the mass spectrometry is used for its identification (Sahil *et al.*, 2011).

Chionanthus mala-elengi (Dennst.) P. S. Green belongs to the family Oleaceae (Jose and Alvaro, 2016). It is locally known as Kallidala and mala-elengi. It is a near threatened endemic tree species in the Peninsular India. It is also distributed in Burma, Thailand, Vietnam, Cambodia and Malaysia (Narayanan *et al.*, 2018; Kiew, 1998; Pius *et al.*, 2015). In folk medicine, leaves and bark of *C. mala-elengi* were used for treating giddiness, epilepsy, wound healing and liver disease (Manilal and Remesh, 2010; Kumar *et al.*, 2016). Yet, no GC-MS studies on stem bark of *C. mala-elengi* have been reported. Hence, the present study was aimed to investigate the chemical constituents in the stem bark ethanolic extract of *C. mala-elengi* through GC-MS analysis.

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Materials and Methods

Plant sample

The stem barks of *C. mala-elengi* were collected from Anaikatti region in Coimbatore district between September 2018 and April 2019. The plant was identified and authenticated by Botanical Survey of India, Coimbatore (BSI/SRC/5/23/2018/Tech/1850).

Sample preparation and extraction

The stem bark of *C. mala-elengi* was used in the study. The collected plant samples were gently washed in tap water, shade dried under room temperature for 3–4 weeks. The dried plant materials were ground to a coarse powder. The powdered plant material (30 g) was extracted with ethanol (150 ml) in Soxhlet apparatus. The mixture was filtered, the filtrates were evaporated and the dried extract was stored in a deep freezer for further use.

Table 1 : GC-MS spectral analysis of stem bark ethanolic extract of *C. mala-elengi*.

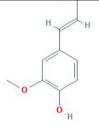
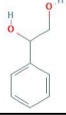
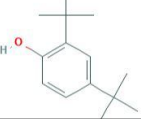
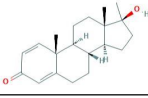
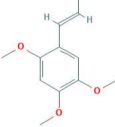
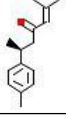
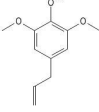
No.	Phytochemical Name	Retention Time(min)	Area%	Molecular weight(g/mol)	Chemical formula	Chemical structure
1	2-(4'-methoxyphenyl)-2-(2'-methoxyphenyl) propane	11.336	1.59	Unkown	Unknown	Unknown
2	(Z)-Isoeugenol	12.127	5.01	164.204	C ₁₀ H ₁₂ O ₂	
3	1-Phenylethylene Glycol	12.617	1.91	138.16	C ₈ H ₁₀ O ₂	
4	2,4-Ditert-Butylphenol	12.681	2.62	206.32	C ₁₀ H ₁₂ O ₂	
5	1,2-Dehydro-17-methyl testosterone	13.855	3.55	300.4	C ₂₀ H ₂₈ O ₂	
6	Alpha-asarone	13.928	5.05	208.25	C ₁₀ H ₁₂ O ₂	
7	Ar-tumerone	14.587	2.40	216.32	C ₁₅ H ₂₀ O	
8	Methoxyeugenol	15.108	3.64	194.18	C ₁₁ H ₁₄ O ₃	

Table 1 contd.....

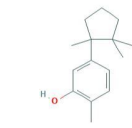
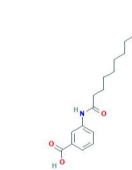
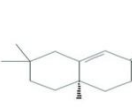
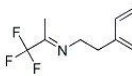
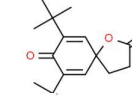
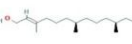
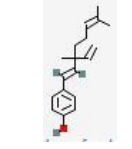
GC-MS analysis

GC-MS analysis of bioactive compounds in stem bark ethanolic extract of *C. mala-elengi* was performed at Kerala Forest Research Institute, Thrissur District, Kerala (Casuga *et al.*, 2016).

Results and Discussion

The scientific validation and extraction of plant materials play an important role in the development of quality control of herbal drugs. Hence, the present investigation was undertaken to find out the therapeutic constituents present in the ethanolic extract of stem bark of *C. mala-elengi* by using Gas chromatography and Mass spectroscopy. The active principles with their Retention time (min), Area%, Molecular weight (g/mol), Chemical formula and Chemical structure were presented in Table. 1 and Fig. 1 which reveals the presence of 16

Table 1 contd.....

No.	Phytochemical Name	Retention Time(min)	Area %	Molecular weight(g/mol)	Chemical formula	Chemical structure
9.	Phenol,2-methyl-5-(1,2,2-trimethylcyclopentyl)-, (S)-	15.596	12.26	218	C ₁₅ H ₂₂ O	
10	3-(Dodecanoylamino)benzoic acid	15.896	24.87	319	C ₁₉ H ₂₉ NO ₃	
11	2(3H)-Naphthalenone, 4,4A,5,6,7,8,-Hexahydro-4A,7,7-trimethyl-, (R)-	16.00	14.27	192.3	C ₁₃ H ₂ O	
12	1H-Inden-1-one,3A,4,5,6,7,7A-hexahydro-5,5-dimethyl-, CIS-	16.100	14.73	Unknown	Unknown	Unknown
13	Benzeneethanamine,N-(2,2,2-Trifluoro-1-methylethylidene)-	16.275	2.71	215.21	C ₁₁ H ₁₂ F ₃ N	
14	7,9-Di-tert-butyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione	17.857	0.88	276	C ₁₇ H ₂₄ O ₃	
15	Phytol	22.311	1.81	296.5	C ₂₀ H ₄₀ O	
16	Phenol,4-(3,7-dimethyl-3-ethenylocta-1,6-dienyl)-	22.623	2.71	256.383	C ₁₈ H ₂₄	

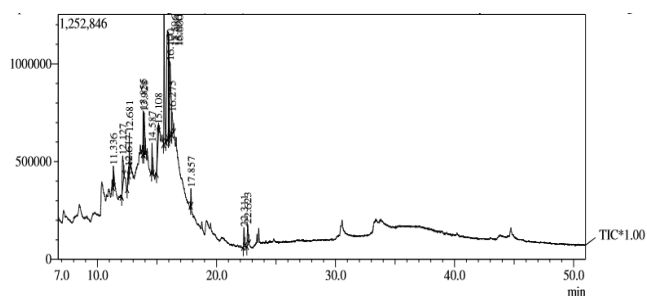


Fig. 1 : GC-MS chromatogram for stem bark ethanolic extract of *C. mala-elengi*.

bioactive compounds in the ethanolic extract of *C. mala-elengi* stem bark. The mass spectra of identified compounds from stem bark ethanolic extract of *C. mala-elengi* were presented in Fig. 2A-2P. Among the identified bioactive compounds, Isoeugenol a phynyl propene, occurs in essential oil of *Ylang ylang* and is synthesized from eugenol. It is used in manufacturing of perfumeries, flavours, essential oil and vanillin. It has

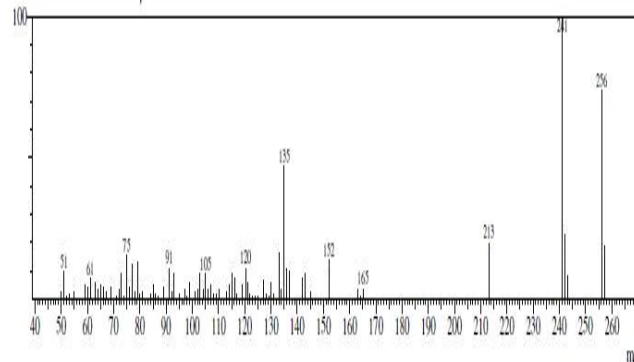


Fig.2:(A): 2-(4'-Methoxyphenyl)-2-(2'-methoxyphenyl)propane.

medicinal properties such as antiseptic and analgesic (Vijay *et al.*, 2017). 2,4-di-tert-butyl phenol (2, 4, DTBP) is a volatile phenolic compound. It has been reported to be present in fruits and seeds. It possess antifungal, antioxidant, antimalarial activities and is also cytotoxic against mammalian cancer cell lines such as H9C2, HeLa and MCF-7. It is also used in pharmaceutical industries

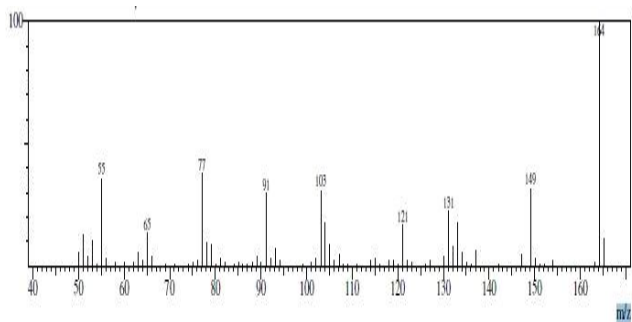


Fig. 2: (B): (Z)-Isoeugenol.

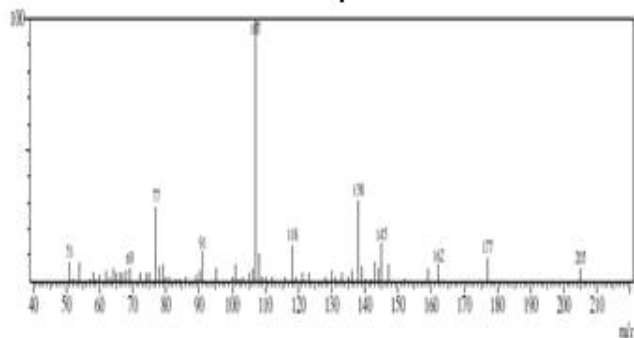


Fig. 2: (C): 1-Phenylethylene glycol.

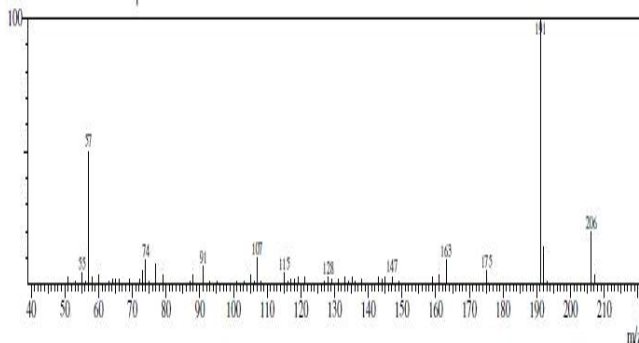


Fig. 2: (D): 2,4-Ditert-butylphenol.

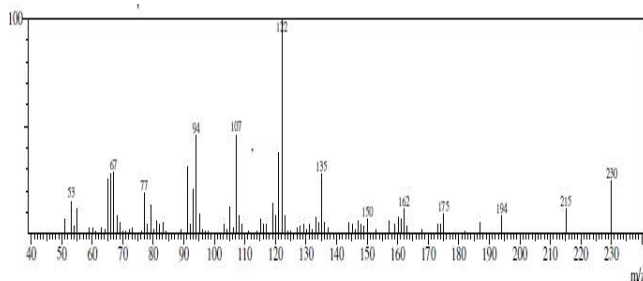


Fig. 2: (E): 1,2-Dehydro-17-methyltestosterone.

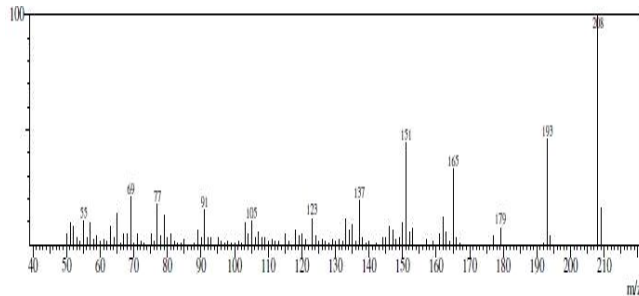


Fig. 2: (F): Alpha-asarone.

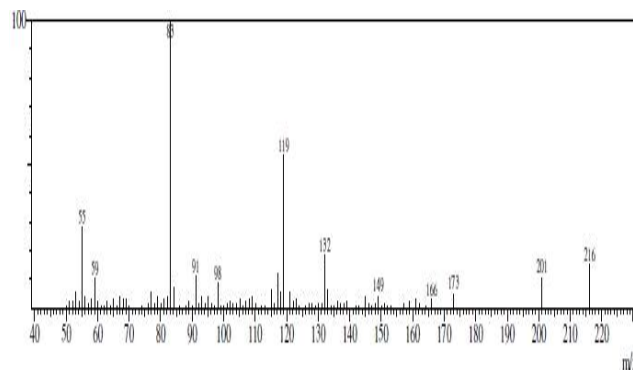


Fig. 2: (G): Ar-turmerone.

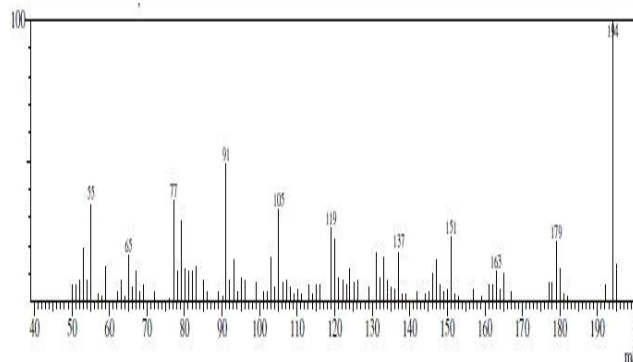


Fig. 2: (H): Methoxyeugenol.

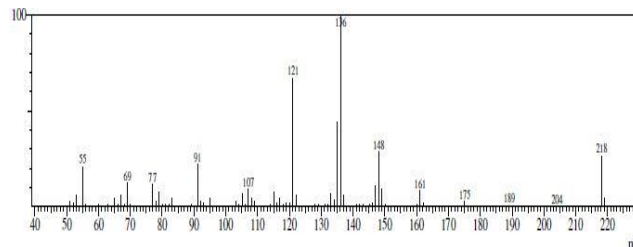


Fig. 2: (I): Phenol, 2-methyl-5-(1,2,2-trimethylcyclopentyl)-, (S)-

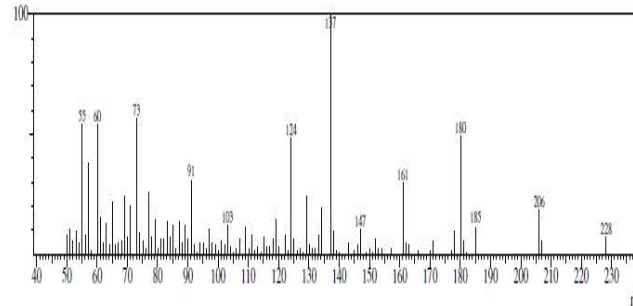


Fig. 2: (J): 3(Dodecanoylamino)benzoic acid.

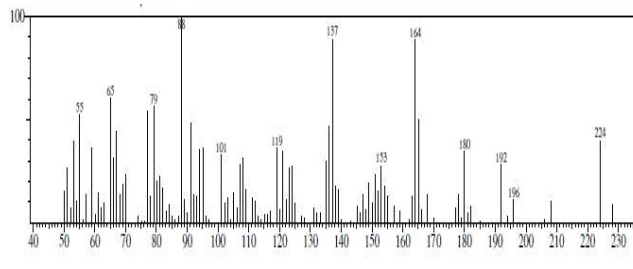


Fig. 2: (K): 2(3H)-Naphthalenone, 4,4a,5,6,7,8-hexahydro-4a,7,7-trimethyl-, (R)-

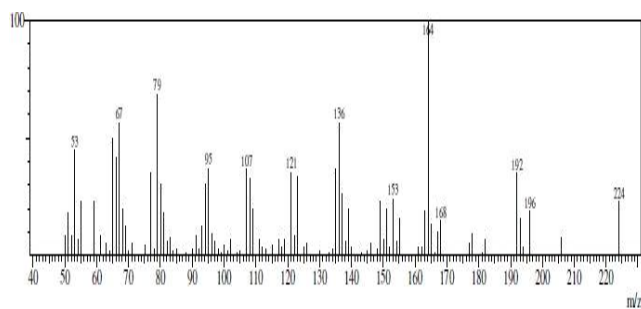


Fig. 2: (L): 1H-Inden-1-one, 3A,4,5,6,7,7A-hexahydro-5,5-dimethyl-, CIS-

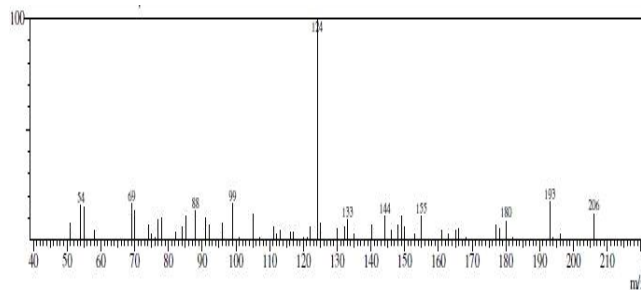


Fig. 2: (M): Benzeneethanamine, N-(2,2,2-trifluoro-1-methylethylidene)-

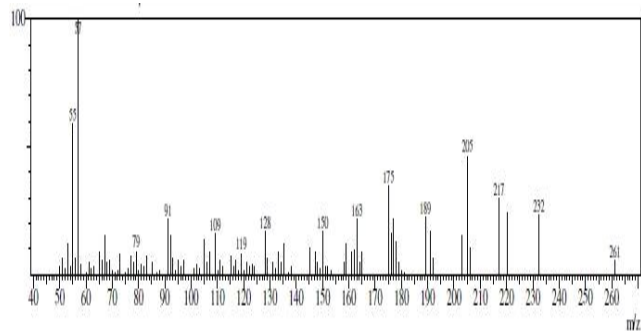


Fig. 2: (N): 7,9-Di-tert-butyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione

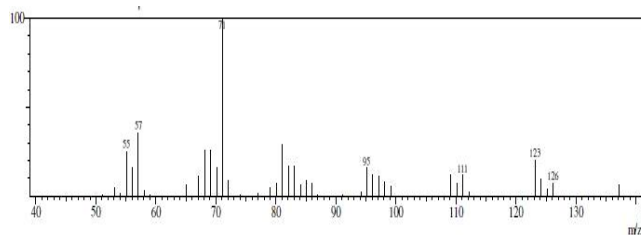


Fig. 2: (O): Phytol

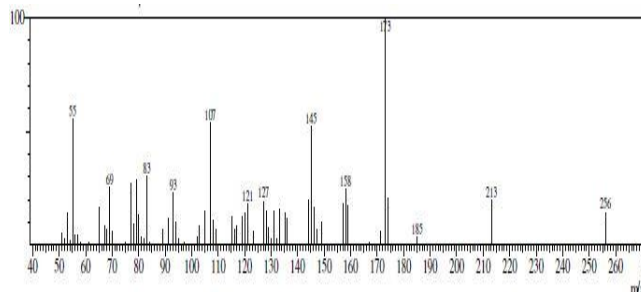


Fig. 2: (P): Phenol, 4-(3,7-dimethyl-3-ethenylocta-1,6-dienyl)-

and manufacturing fragrances (Varsha *et al.*, 2015). 7, 9-Di-tert-butyl-1-oxaspiro (4, 5) deca-6, 9-diene-2, 8-dione has antioxidant property (Chandrasekar *et al.*, 2015).

Alpha-asarone is one of the Psychoactive phytochemical. It has numerous pharmacological activities such as anticonvulsant, antidepressant, anti-anxiety, anti-alzheimer, antiparkinson, antiepileptic, anticancer, antihyperlipidemic, antithrombotic and anticholestatic effects (Chellian and Pandey, 2018; Chellian *et al.*, 2017). Phytol exhibits antioxidant and antinociceptive effects. Phytol, a precursor of synthetic vitamin E and vitamin K, was proven to be cytotoxic against breast cancer cell lines (MCF7) (Casuga *et al.*, 2016). Ar-tumerone is effective in treating neurodegenerative disease and stroke. It is also responsible for the antitumor properties, anti-inflammatory properties, apoptosis, inhibition of tumor cell invasion, parkinson disease and alzheimer disease. It is cytotoxic activity to HL-60, K-562, L-1210, Hela U-937 and RBL-2H3 cells (Hucklenbroich *et al.*, 2014; Essien *et al.*, 2015).

Conclusion

C. mala-elengi is a natural plant of Peninsular India. Qualitative determination of ethanolic stem bark extract of *C. mala-elengi* using Gas chromatography mass spectrometry revealed the presence of various bioactive compounds. The results of the study concluded that the most of the compounds possess considerable antioxidant potential. Based on this property, further studies will be carried out through in vivo assays of ethanolic stem bark extract of *C. mala-elengi*.

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