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Research Article

Determination of Bioactive Components of *Asystatia travancorica* Bedd (Acanthaceae) by GC-MS Analysis.

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ABSTRACT
In this study, the bioactive compounds of whole plant of *Asystasia travancorica* have been evaluated using GC-MS analysis. The chemical compositions of the ethanol extract of whole plant of *Asystasia travancorica* was investigated 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 Standard and Technology (NIST) library. Ten compounds were identified. This analysis revealed the presence of Levo-a-Elemene (41.97 %), Tetrahydtospirilloxanthin (17.07%), Stigmasterol (12.25%), Phytol (8.23%), 2,6,10-Dodecartrien-1-ol, 3,7,11-trimethyl-[trans-farnesol] (7.03%) and Ethyl iso-allochote (4.62%).

Keywords: *Asystasia travancorica*, GC-MS analysis, Phytocomponents, Stigmasterol

INTRODUCTION

Plants produce a remarkable diverse array of over 5, 00,000 low and high molecular mass natural products which are known as secondary metabolites [(1)]. Distinguished example of these compounds includes flavonoids, phenols, saponins and cyanogenic glycosides [(2,3)]. It has been shown that *in vitro* screening methods could provide the needed preliminary observations necessary to select crude plant extracts with potentially helpful properties for further chemical and pharmacological investigations [(4)]. Natural products from microbial sources have been the primary source of antibiotics, but with the increasing recognition of herbal medicine as an alternative form of healthcare, the screening of medicinal plants for active compounds has become very significant [(5)].

Asystasia includes approximately 70 species of perennial herbs and subshrubs from tropical Africa, India and Asia. *Asystasia* belongs to the family Acanthaceae. Paste of leaves and flowers of *A.travancorica* mixed with honey is taken orally, twice a day, for three weeks for the treatment of rheumatism [(6)]. To our knowledge, no chemical analysis has been previously reported on this plant. The present communication deals with the GC-MS analysis of ethanol extract of whole plant of *A.travancorica*.

MATERIALS AND METHODS

Collection of Plant Sample: Whole plant of *A.travancorica* was collected from Agasthiarmalai Biosphere Reserve, Western Ghats, Tamil Nadu with the help of local flora. The specimens were identified and preserved in the Ethnopharmacology Unit, Research Department of Botany, V.O. Chidambaram College, Tuticorin, Tamil Nadu.

Preparation of Plant Extract: The whole plant of *A.travancorica* was cleaned, shade dried and pulverized to powder in a mechanical grinder. Required quantity of powder was weighed and transferred to stoppered flask, and treated with ethanol until the powder is fully immersed. The flask was shaken every hour for the firsts 6 hours and then it was kept aside and again shaken after 24 hours. This process was repeated for 3 days and then the extract was filtered. The extract was collected and evaporated to dryness by using vacuum distillation unit. The final residue thus obtained was then subjected to GC – MS analysis.

GC-MS Analysis: GC-MS analysis of ethanol extract was performed with GC clarus 500 Perkin Elmer system and Gas chromatograph interfaced to a Mass spectrometer (GC-MS) equipped with a Elite – 1 fused silica capillary column (30 mm x 0.25 mm 1D x 1 um df, composed of 100% Dimethyl poly siloxane). For GC-MS detection, and electron ionization system with ionizing energy of 70 eV was used. Helium gas (99.999%) was used as the carrier gas at constant flow rate 1 ml / min and an injection volume of 2 ul was employed (Split ratio of 10:1); Injector temperature 2500C; ion-source temperature 2800C. The oven temperature was programmed from 1100 C (isothermal for 2 min) with an increase of 100C/min, to 2000C, then 50C/min to 2800C, ending with a 9 min isothermal at 2800C. Mass spectra were taken at 70 eV, a scan interval of 0.5 seconds and fragments from 45 to 450 Da. Total GC running time was 36 minutes. The relative % amount of each component was calculated by comparing

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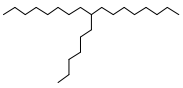
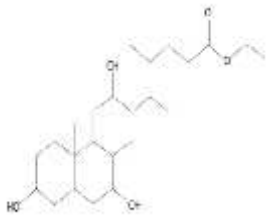
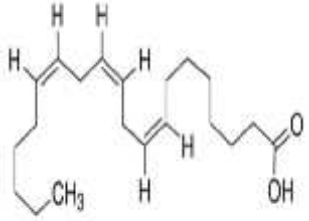
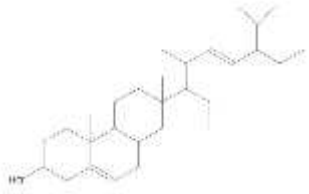
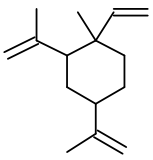
Table 1: Phytocomponents detected in *Asystasia travancorica* whole plant

SI No	RT	Name of Compound	Molecular Formula	Molecular Weight	Peak Area %	Structures
1	5.49	Heptadecane, 9-hexyl-	$C_{23}H_{48}$	324	3.21	
2	8.08	Tetratetracontane	$C_{44}H_{90}$	618	1.41	
3	11.45	Ethyl iso-allocholate	$C_{26}H_{44}O_5$	436	4.62	
4	13.24	Palmitic acid, ethyl ester	$C_{18}H_{36}O_2$	284	2.21	
5	14.72	Phytol	$C_{20}H_{40}O$	296	8.23	
6	15.46	8,11,14-Eicosatrienoic acid, methyl ester, (Z,Z,Z)- [Methyl dihomo-ç-linolenate]	$C_{21}H_{36}O_2$	320	2.01	
7	24.31	2,6,10-Dodecatrien-1-ol, 3,7,11-trimethyl-, (E,E)- [trans-Farnesol]	$C_{15}H_{26}O$	222	7.03	
8	30.52	Stigmasterol	$C_{29}H_{48}O$	412	12.25	
9	31.74	Tetrahydrospirilloxanthin	$C_{42}H_{64}O_2$	600	17.07	
10	33.57	levo-á-Elemene	$C_{15}H_{24}$	204	41.97	

its average peak area to the total areas, software adopted to handle mass spectra and chromatograms was a turbomass. Interpretation on mass spectrum of GC-MS was done using the database of National Institute of Standard and Technology (NIST) having more than 62,000 patterns. The mass 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.

RESULT AND DISCUSSION

The results pertaining to the GC-MS analysis are presented in Figure-1 and Table-1.Ten compounds were detected in ethanol extract of whole plant of *A.travancorica*. The

Table.2 Activity of phytocomponents identified in the ethanol extract of *Asystasia travancorica* whole plant

S.No.	Name of the compound	Molecular formula	Compound nature	**Activity
1.	Ethyl iso-allocholate	$C_{26}H_{44}O_5$	Steroid	Antimicrobial Anti-inflammatory Diuretic Antiasthma Antiarthritic Anti cholesterol
2.	Palmitic acid, ethyl ester	$C_{18}H_{36}O_2$	Fatty acid ester	Antioxidant, Hypocholesterolemic Nematicide, Pesticide, Lubricant, Antiandrogenic, Flavor, Hemolytic 5-Alpha reductase inhibitor
3.	Phytol	$C_{20}H_{40}O$	Diterpene	Diuretic Anticancer Anti-inflammatory Antimicrobial
4.	2,6,10-Dodecatrien-1-ol, 3,7,11-trimethyl-, (E,E)-[trans-Farnesol]	$C_{15}H_{26}O$	Sesquiterpene alcohol	Antimicrobial Anti-inflammatory
5.	Stigmasterol	$C_{29}H_{48}O$	Phytosterol	Antimicrobial Anti-inflammatory Diuretic Antiasthma Antiarthritic Anti cholesterol
6.	Tetrahydrospirilloxanthin	$C_{42}H_{64}O_2$	Carotenoid	Antioxidant Anti-inflammatory Antiarthritic Lowering hypertension Anticancer Drug for eye problems Natural color pigment
7.	levo-á-Elemene	$C_{15}H_{24}$	Sesquiterpene	Anti-tumor, Fungicide Analgesic, Antibacterial, Anti-inflammatory, Sedative

***Source: Dr.Duke's Phytochemical and Ethnobotanical Databases*

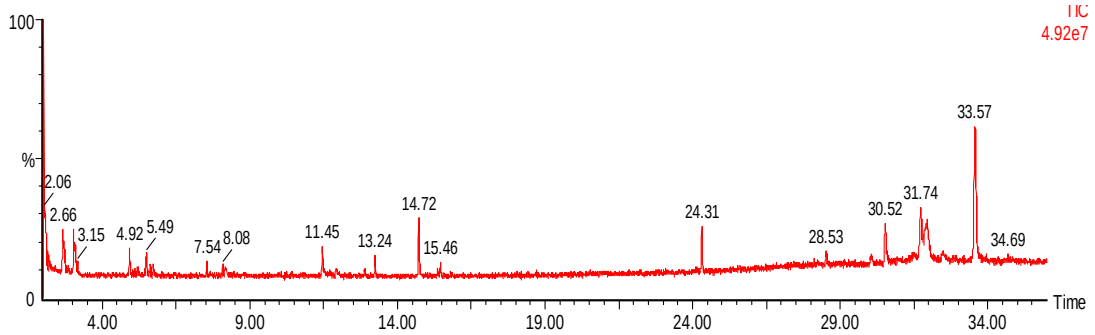


Fig 1: GC-MS chromatogram of the ethanol extract of Asystasia travancorica whole plant

results revealed that, Levo-a-Elemene (41.97%), Tetrahydrospirilloxanthin (17.07%), Stigmasterol (12.25%), Phytol (8.23%), 2,6,10 – Dodecartrien -1-ol, 3,7,11-trimethyl- [trans-farnesol] (7.03%), Ethyl iso-allocholate (4.62%) and Heptadecane 9-hexyl (3.21%) were found as the major components in the ethanol extract of *A.travancorica*. Very small quantity of palmitic acid, ethyl ester (2.21%), 8,11,14, Eicosatrienoic acid, methyl ester, (z,z,z) – [methyl dihomo- c- linolenate] (2.01%) and Tetratetracontane (1.41%) were reported.

Table-2 listed various phytochemical constituents which contribute to the medicinal activity of ethanol extract of whole plant of *A.travancorica*. Among the identified phytocochemicals, stigmasterol is used as a precursor of semisynthetic progesterone [(7)], a valuable human hormone that plays an important physiological role in the regulatory and tissue rebuilding mechanisms related to estrogens effect, as well as acting as an intermediate in the biosynthesis of androgens, estrogens and corticoids. It is also used as the precursor of Vitamin D_3 [(8)]. Phytol is one among the ten compounds of the present study. 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 [(9)]. Phytol is a key acyclic diterppene alcohol that is a precursor for Vitamins E and K_1. It is used along with simple or corn syrup as a hardener in candies. Phytol is detected in whole plant of *A.travancorica* which was also found to be effective at different stages of the arthritis. It was found to give good as well as preventive and therapeutic results against arthritis [(10)].

In the present study, ten components from whole plant of *A.travencorica* were identified by GC – MS analysis. The presence of various bioactive compounds justifies the use of this plant for various ailments by traditional practitioners. So that it might be utilized for the development of traditional medicines and further investigation is in need to elute novel active compounds from the medicinal plants which may create a new way to treat many incurable diseases.

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