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GC-MS ANALYSIS OF BIOACTIVE COMPONENTS OF AN IMPORTANT MEDICINAL FERN *ACTINIOPTERIS RADIATA* (SWARTZ) LINK.

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ABSTRACT

The phytochemical constituents are responsible for medicinal activities of plant species. The present investigation was carried out to analyze the bioactive components from the whole plant of *Actiniopteris radiata* (Pteridaceae) using GC-MS technique. The chemical compositions of the ethanolic extract of *A. radiata* were investigated using Perkin-Elmer Gas Chromatography–Mass Spectrometry and five bioactive phytochemical compounds were identified. The ethanolic extract of *A. radiata* revealed the existence of Hexadecanoic acid, ethyl ester (20.40%), 9,12-Octadecadienoic acid, methyl ester, (E,E)- (10.91%), (E)-9-Octadecenoic acid ethyl ester (63.59%), Docosanoic acid, ethyl ester (4.26%), Heptadecanoic acid, heptadecyl ester (0.84%). The present article revealed the detailed exploration of phytoconstituents and pharmacological activities and confirms the application of *A. radiata* for various ailments. In future, the isolation of individual phytochemical compound may lead to find a novel drug.

KEYWORDS: *Actiniopteris radiat*a, Pteridaceae, GC-MS analysis, bioactive components, ethanolic extract.

I. INTRODUCTION

Plants are indispensable to human beings for their entire life. The history of herbal medicine starts from the ancient human civilization. The wealth of India is stored in the enormous natural flora which has been gifted to Indians. The phytochemistry has been studied in less number of non-flowering plants in contrast to flowering plants. Phytochemistry is one of the more fashionable and rapidly expanding areas of plant taxonomy (chemosystematics) which

utilizes chemical information to improve the classification of plants. The phytochemical characters could be used as markers to identify and differentiate the species. Phytochemical analysis of ferns makes the basis for the investigations on medicinal uses of the plants.[1] The secondary metabolites from natural products are showing more drug likeness and biologically friendliness than total synthetic molecules. In earlier days drug targets were exposed to crude extracts. But now a day the extracts were fractionated, active compounds isolated and characterized.[2]

A. radiata (Swartz) Link. belongs to the family Pteridaceae. It is commonly called Peacock's tail having herbaceous miniature palm like fronds and lithophytic in its distribution. In Tamil it is called as Mayilatum shikhai. The plants are 8-25cm height, rooting in the crevices of rock or moist and shady places. The rhizome is oblique to horizontal, 1.5 to 2.0cm in length, densely covered by scales and leaf bases. The young fronds show circinate venation and lamina flabellate, semicircular or wedge-shaped, dichotomously divided into linear segments and dimorphic with fertile and sterile fronds. Usually fertile fronds are larger than the sterile fronds. Sori are arranged in two rows on the lower side of the pinnae lobes. The sporangia borne in intra-marginal grooves throughout, protected by the reflexed margin of the segment.

The plant is bitter, astringent, sweet, cooling, acrid, constipating, anthelmintic, haemostatic, antileprotic and febrifuge. It is useful in vitiated conditions of kapha and pitta, diarrhoea, dysentery, leprosy, skin diseases, diabetes and fever. The Literature reported that the traditional medicinal preparations from whole plant have long been used as folklore medicine for treatment of various diseases.[3,4] The plant is also known for its rich primary and secondary metabolites.[5] Some work was carried out in the crude drug of this plant for its pharmacological activity.[6,7,8] Hence the present study is carried out to find out the bioactive chemical constituents from the ethanolic extract of whole plant of *A. radiat*a by Gas Chromatogram Mass spectrometry (GC-MS) analysis which is an advanced and accurate one to find out the bioactive compounds.

II. MATERIALS AND METHODS

Collection

A. radiata is commonly found in hilly areas or Western Ghats. It is located at 10.12Â° N 77.55Â° E. It has an average elevation of 282 meters (925 feet). It is located at the foothills of the Western Ghats. It is one of the most fertile places in the state of Tamil Nadu. The plant materials were identified from The Rapinat Herbarium and Center for Molecular Systematics,

St. Joseph's College, Tiruchirappalli, Tamil Nadu, India. The voucher specimens were deposited in the Department of Botany, Holy Cross College (Autonomous), Tiruchirappalli for future reference.

Sampling of Plant material for GC-MS analysis

10gm of powdered whole plant material was soaked in 20ml of absolute alcohol overnight and then filtered through Whatmann No.1 filter paper along with 2gm Sodium sulfate to remove the sediments and traces of water in the filtrate. Before filtering, the filter paper along with sodium sulphate was wetted with absolute alcohol. The filtrate is then concentrated by bubbling nitrogen gas into the solution and was concentrated to 1ml. The extract contains both polar and non-polar phytocomponents.

GC-MS Analysis

GC-MS analysis was carried out on a GC Clarus 500 Perkin Elmer system and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following conditions: Column Elite-5MS fused silica capillary column (30mm×0.25mm ID ×1 μmdf, composed of 5% Diphenyl / 95% Dimethyl poly siloxane), operating in electron impact mode at 70 eV; Helium (99.999%) was used as carrier gas at a constant flow of 1ml/min and an injection volume of 2 μl 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 45 to 450 Da. Total GC running time was 36 min. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. The mass-detector used in this analysis was Turbo-Mass Gold-Perkin-Elmer, and the software adopted to handle mass spectra and chromatograms was a Turbo-Mass ver-5.2.

Identification of Components[9]

Interpretation on mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology (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.

III. RESULTS

The studies on the bioactive components in the ethanolic extract of whole plant of *A. radiata* by GC-MS analysis clearly showed the presence of five bioactive compounds. The active principles with their retention time (RT), molecular formula (MF), molecular weight (MW) and concentration (peak area %) were presented in Table-1. The GC-MS chromatogram of the five peaks of bioactive compounds detected were shown in Figure-1. There were five active phytoconstituents identified by the mass spectroscopy. The total numbers of compounds identified in ethanolic extracts were Hexadecanoic acid, ethyl ester (20.40%), 9,12-Octadecadienoic acid, methyl ester, (E,E)- (10.91%), (E)-9-Octadecenoic acid ethyl ester (63.59%), Docosanoic acid,ethyl ester (4.26%), Heptadecanoic acid, heptadecyl ester (0.84%). Fig-2, 3, 4, 5 and 6 showed the mass spectrum of five bioactive constituents.

Table-1: Components detected in ethanolic extract of *A. radiata* (Swartz) Link.

S.No	Retention Time	Name of the Compound	Molecular Formula	Nature of the compound	Molecular Weight	Peak Area
1	12.81	Hexadecanoic acid,ethyl ester	$C_{18}H_{36}O_2$	Palmitic acid ester	284	20.40
2	14.88	9,12-Octadecadienoic acid, methyl ester, (E,E)-	$C_{19}H_{34}O_2$	Linoleic acid ester	294	10.91
3	14.97	(E)-9-Octadecenoic acid ethyl ester	$C_{20}H_{38}O_2$	Oleic acid ester	310	63.59
4	15.29	Docosanoic acid, ethyl ester	$C_{24}H_{48}O_2$	Fatty acid ester	368	4.26
5	18.0	Heptadecanoic acid, heptadecyl ester	$C_{34}H_{68}O_2$	Fatty acid ester	508	0.84

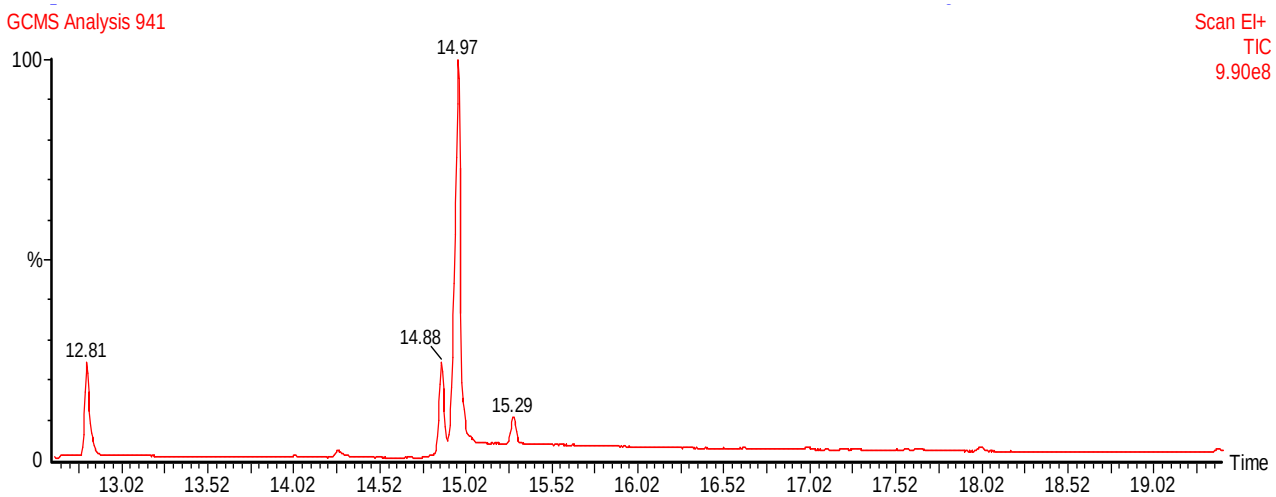

Fig-1: Chromatogram obtained from GC-MS with the ethanolic extract of *A. radiata* (Swartz) Link.

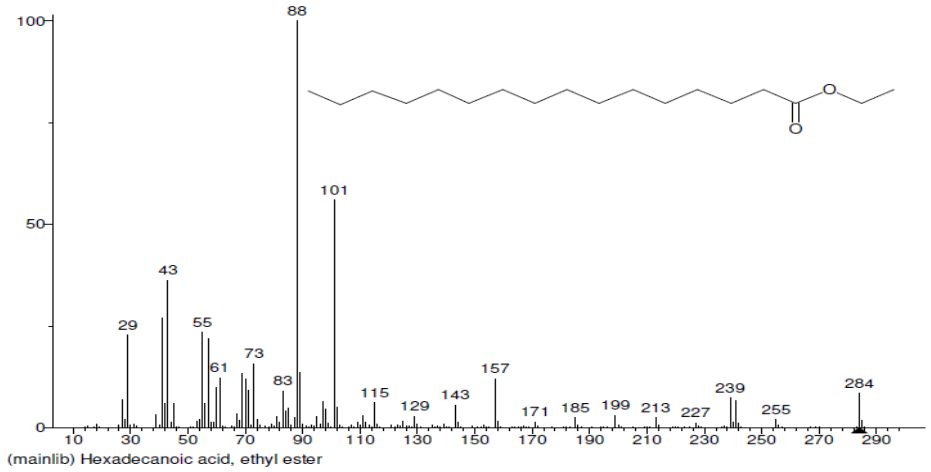

Fig-2

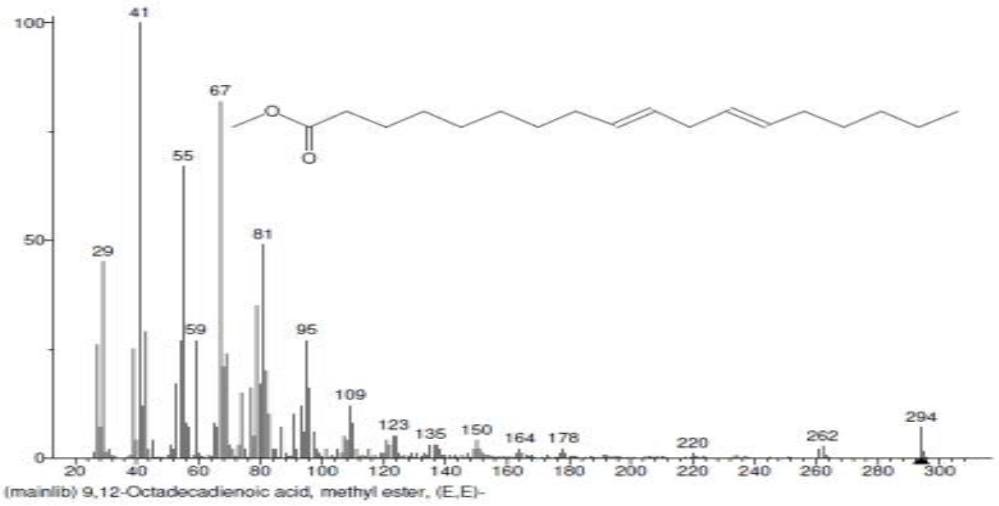

Fig-3

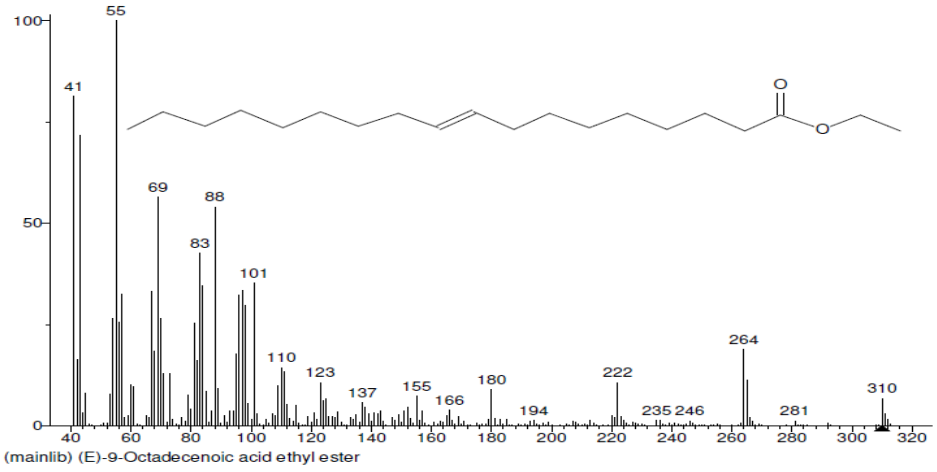

Fig-4

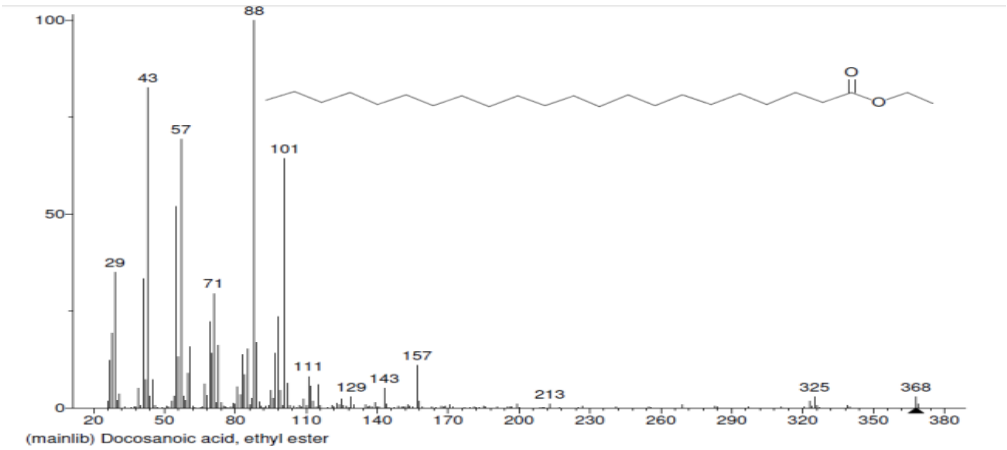

Fig-5

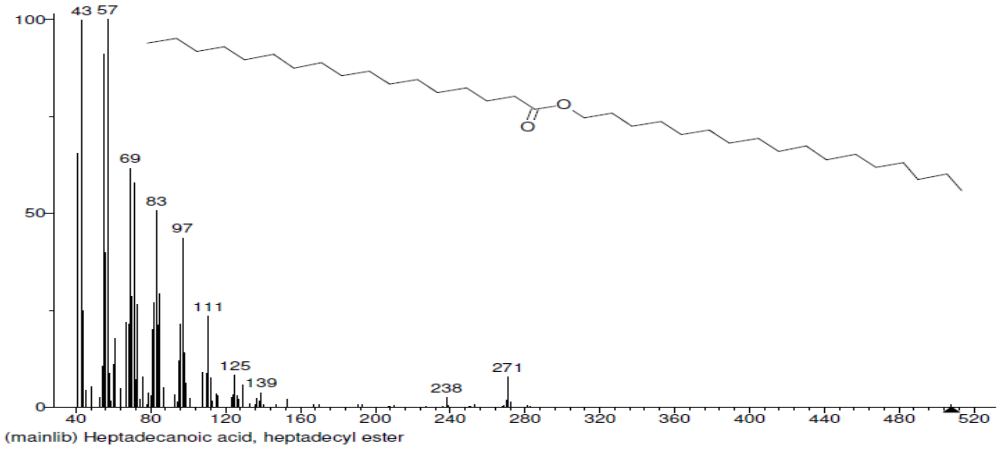

Fig-6

IV. DISCUSSION

In the analysis of bioactive components of whole plant of *A. radiata* through GC-MS showed the presence of five bioactive compounds such as Hexadecanoic acid, ethyl ester, 9,12-Octadecadienoic acid, methyl ester, (E,E)-, (E)-9-Octadecenoic acid ethyl ester, Docosanoic acid,ethyl ester and Heptadecanoic acid, heptadecyl ester. Hexadecanoic acid, ethyl ester showed the antioxidant, hypocholesterolemic, lubricant, antiandrogenic, hemolytic 5-Alpha reductase inhibitor activity. The second compound such as 9,12-Octadecadienoic acid, methyl ester, (E,E) showed the activity of cancer preventive, hypocholesterolemic, hepatoprotective, nematicide, insectifuge, antihistaminic, antieczemic, antiacne, alpha reductase inhibitor, antiandrogenic, antiarthritic and, anti-inflammatory. The third compound such as (E)-9-Octadecenoic acid ethyl ester showed anti-inflammatory, cancer preventive, dermatitigenic hypocholesterolemic, 5-Alpha reductase inhibitor and antiandrogenic activity.

These biological activities were obtained based on Phytochemical and Ethnobotanical Databases.[10]

The present GC-MS report was supported by many authors in some Pteridophytes. The bioactive compound such as hexadecanoic acid has the property of antioxidant activity[11] and hexadecanoic acid has the property of hypochloresterolenic, nematiside, pestiside, lubricant, anti androgenic flavor and hemolytic properties.[12] The Oleic acid, hexadecanoic acid, octadecanoic acid are known to have potential antibacterial and antifungal activity.[13,14]

Our results are co incide with the reports of some authors. They reported the activities of some plant constituents with compounds nature of flavonoids, palmitic acid (hexadecanoic acid, ethyl ester and hexadecaonoic acid), unsaturated fatty acid and linolenic (docosatetraenoic acid and octadecatrienoic acid) as antimicrobial, anti-inflammatory, antioxidant, hypocholesterolemic, cancer preventive, hepatoprotective, antiarthritic, antihistimic, antieczemic and anticoronary.[15] The results of GC-MS analysis of various extracts of *Clerodendrum phlomidis* leaf and methanolic extract of *Woodfordia fruticosa* revealed the presence of twenty one compounds.[16,17]

The ethanolic extract of *Polygonum chinense* revealed the presence of five phytochemical constituents and reported as having antimicrobial activity.[18] In *Polypodium decumanum*, 13 individual compounds were identified.[19] The compound unsaturated fatty acid and octadec-9-enolic acid were identified and explained that the compounds lowers the blood level of cholesterol and lowers the risk of heart problem and also responsible for hypotensive, atherosclerosis and aids in cancer prevention.[20] The natural product pterosin was identified in the fern *Pteridium aquilinum* and reported to exhibit smooth muscle relaxant properties.[21]

The essential oil composition of the aerial parts of aromatic fern *Anemia tomentosa* var. *anthriscifolia* through GC-MS was characterized and explained. The volatile oil was composed mainly of sesquiterpenes with lower amounts of monoterpenes. The sesquiterpenes were composed mainly of oxygenated components (67%), including α-bisabolol (51%), spathulenol (1%), caryophyllene oxide (3%), α-bisaboloxide (1%) and 14-hydroxy-9-epi-(*E*)-caryophyllene (1%). The monoterpenes were dominated by neral (5%) and geranial (7%), with lower amounts of α-pinene, camphene, 6-methyl-5-hepten-2-one, 1, 8-cineole and pinocarveol.[22] A new triterpenoid, 22, 29-xi-epoxy-30-norhopane-13beta-ol isolated from *Adiantum lunulatum*.[23]

V. CONCLUSION

In the present study five bioactive chemical constituents have been identified from ethanolic extract of whole plant of *A. radiat*a by Gas Chromatogram Mass spectrometry (GC-MS) analysis. The above said bioactive compounds found in this fern are being used for the pharmacological work. Thus this type of GC-MS analysis is the first step towards understanding the nature of active principles in the folklore medicine. However, the isolation of individual phytochemical constituent and subjecting it to biological activity will definitely give fruitful results. It could be concluded that, the presence of various bioactive compounds justifies the whole plant is used as a remedy for various ailments by traditional practitioners. So it is recommended as plant of pharmaceutical importance. However, further studies are needed to undertake its bioactivity and toxicity profile.

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