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Research Article

INVESTIGATION OF CHEMICAL COMPOSITION FROM *DRYOPTERIS CHOCHLEATA* (D. DON) C. CHR. (DRYOPTERIDACEAE)

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ABSTRACT

Objective: The present study was for the determination of bioactive volatile compounds from chloroform and ethanolic extract.

Methods: For present investigation of the samples were carried out by using Shimadzu Make QP-2010 with nonpolar 60 M RTX 5MS Column. Interpretation on Mass-Spectrum GC-MS was conducted using the database of National Institute Slandered and Technology (NIST) having more 62,000 patterns.

Results: In Ethanolic extract seven compounds and chloroform extract fourteen compounds were identified. The common compounds in these two extracts are Germacrene D; 1, 3-cyclohexanedione, 2-methyl-2 (3-Oxobutyl); Neoisolongifolene, 8, 9-dehydro.

Coclusion: Present investigation emphasizes the efficacy of traditional remedies and that it inspires the people to realize the importance of natural resources for its potent pharmaceutical use.

Keywords: Medicinal pteridophyte, Gas chromatography-mass spectroscopy analysis, Ethanol extract, Chloroform extract, Bioactive compounds.

INTRODUCTION

India is the largest producer of medicinal herbs known as the botanical garden of the world [1]. Approximately, 20% of the plants found in the world have been submitted to pharmaceutical or biological tests [11]. Use of plant as a source of medicine has been inherited as an important component of the healthcare system. They are capable of synthesizing an overwhelming variety of low-molecular weight organic compounds called as secondary metabolites, usually with unique complex structures. Many metabolites have been found to possess interesting biological activities and find applications in pharmaceuticals, insecticides, dyes, flavors, and fragrance [3].

The pteridophytes have an important role in traditional medicine. Man has been exploiting pteridophytes prosperity from time immemorial to cure various types of diseases traditionally. These plants have been effectively used in the different systems of medicines such as Ayurvedic, Unani, Homeopathic, Allopathic, Siddha, Naturopathic, home remedies, and other systems of medicines [4]. Many attempts have been made on the study of relationships of pteridophytes with man and more particularly for their medicinal values. *Dryopteris cochleata* (D. Don) C. belongs to the family of Dryopteridaceae, a common, medium-sized terrestrial herb found at higher altitudes in semi-exposed, well-shaded localities in forest, and grasslands [5].

The present work was designed to investigate the chemical profile of *D. cochleata* rhizome. This plant considered to possess potential therapeutic value. Extract of rhizome is used for epilepsy, leprosy, blood purification, as a tonic for strength, for cuts, wounds, ulcers, swellings, pains, etc. It has antifungal properties and also used as an antidote, for snake and dog bites, leaves show antibacterial activity [5,10].

Hence, the present investigation is carried out to determine the possible chemical constituents of *D. cochleata* rhizome extract. In the present study, two different solvents extract of rhizome were analyzed using gas chromatography-mass spectroscopy (GC-MS) technique to study chemical constituents of rhizome.

METHODS

Plant material

The plant namely *D. cochleata* (D. Don) C. was collected from Northern Western Ghats and was identified with the help of floras *viz*., Manickam and Irudayaraj (1992), Fraser-Jenkins (2007), etc. And also personal consult with Rahul Mahamuni.

The collected plant was washed thoroughly with distilled water, separated fronds and rhizome, and subjected to drying in the shadow till proper drying. Thus, dried rhizome was powdered finally using grinder. The sample is transferred into airtight container with proper labeling.

Preparation of plant extract

The plant extract was prepared by Soxhlet extraction method. About 5 g of powder plant material was uniformly packed into a thimble and extracted with ethanol. Extraction continues for 8-9 hrs. After that, the extract was taken in an evaporating dish and heated till all the solvent got evaporated. Dried extract was soared in refrigerator at 4°C for further process. The extract then concentrated to 5 ml and was tested for the presence of bioactive compounds using slandered method.

GC-MS analysis

The rhizome ethanolic and chloroform extract obtained from medicinal plant, i.e., *D. cochleata* (D. Don) C. was subjected to GC-MS for the determination of bioactive volatile compounds. Some of the important features are summarized below. GC-MS analyses of the samples were carried out using Shimadzu make QP-2010 with nonpolar 60 M RTX 5MS column. Helium was used as the carrier gas and the temperature programming was set with initial oven temperature at 400°C and held for 3 minutes and the final temperature of the oven was 4800°C with rate at 100°C [minutes^{-1}]. A 2 µL sample was injected with the splitless mode. Mass spectra were recorded over 35-650 amu range with electron impact ionization energy 70 eV. The total running time for a sample is 45 minutes. The chemical components from the ethanolic extracts of plant were identified by comparing the retention times of chromatographic peaks using Quadra pole detector with National Institute Slandered and Technology (NIST) library to relative retention

indices. Quantitative determinations were made by relating respective peak areas to total ion chromatogram areas from the GC-MS.

Identification of phytocompounds

Interpretation on mass-spectrum GC-MS was conducted using the database of NIST having more 62,000 patterns. The spectrum of the unknown components was compared with the spectrum of 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 phytochemical components of each extract are presented separately in Tables 1 and 2. The GC-MS chromatogram with a peak area of both ethanolic and chloroform rhizome extract is also given in Figs. 1 and 2. Sum total of 21 constituents were identified in the present study from these two solvent extracts.

Chloroform extract recorded the highest number of fourteen compounds. Among these, 9 were most prevailing compounds (Tables 3 and 4). Similarly, in ethanol extract, seven compounds were observed of which 4 compounds were prevailing compounds (Tables 1 and 2). The common compounds in these two extracts are Germacrene D, 1, 3-cyclohexanedione 2-methyl-2 (3-oxobutyl), neoisolongifolene, 8, 9-dehydro. The chromatogram (Fig. 1) of ethanol rhizome extract shows 4 prominent peaks as dimethyl sulfoxide ($C_2H_6O_5$) with retention time 6.911 and peak area of 17.84, neoisolongifolene, 8, 9-dehydro ($C_{15}H_{22}$) with retention time 18.260 and peak area of 2.18, Germacrene D ($C_{15}H_{24}$) with retention time 18.492 and peak area of 15.27, cyclopropane ($C_{18}H_{36}$) with retention time 20.208 and peak area of 13.14. The other less prominent peaks at other retention times are showing the peak identities of the various compounds identified as shown in Fig. 1 and Table 1.

The chromatogram (Fig. 2) of chloroform rhizome extract shows 9 prominent peaks as 3-hexadecene, (Z) ($C_{16}H_{22}$) with retention time 16.825 and peak area of 1.05, neoisolongifolene 8, 9-dehydro ($C_{15}H_{22}$) with retention time 18.483 and peak area of 2.34, Germacrene D ($C_{15}H_{24}$) with retention time 18.483 and peak area of 8.14, 1-pentadecene ($C_{15}H_{30}$) with retention time 19.650 and peak area of 2.58, hexadecane ($C_{16}H_{34}$) with retention time 19.733 and peak area of 0.70, 1-heptadecene ($C_{17}H_{34}$) with retention time 22.133 and peak area of 2.88, 1,2-benzenedicarboxylic acid, bis (2-methylpropyl)

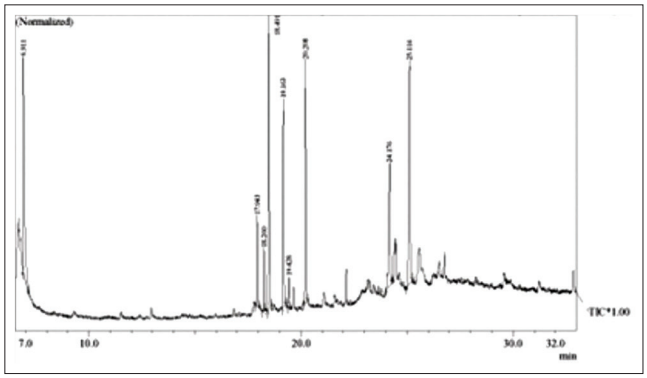


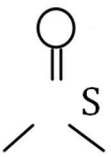
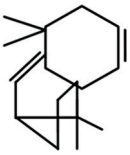
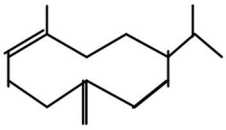
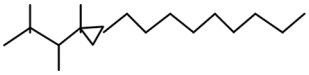
Fig. 1: GC-MS chromatogram of ethanol extract of whole plant extract of *Dryopteris cochleata* (D. Don) C

Table 1: GC-MS analysis revealed the presence of phytochemical components in ethanol rhizome extract of *D. cochleata* (D. Don) C

Peak number	Retention time	Peak area (%)	Name of the compound	Molecular weight	Molecular formula
1	6.911	17.84	Dimethyl Sulfoxide	78	$C_2H_6O_5$
2	17.943	5.53	1,3-cyclohexanedione, 2-methyl-2-(3-oxobutyl)	196	$C_{11}H_{16}O_3$
3	18.260	2.18	Neoisolongifolene, 8, 9-dehydro	202	$C_{15}H_{22}$
4	18.492	15.27	Germacrene D	204	$C_{15}H_{24}$
5	19.167	12.65	Cyclohexane,1,1,3-trimethyl-2-(3-methylpentyl)	210	$C_{15}H_{30}$
6	19.425	1.38	1,6,10-dodecatrien-3-ol, 3,7,11-trimethyl	222	$C_{15}H_{26}O$
7	20.208	13.44	Cyclopropane	252	$C_{18}H_{36}$

GC-MS: Gas chromatography-mass spectroscopy, *D. cochleata: Dryopteris cochleata*

Table 2: Chemical structure of the most prevailing compounds of ethanol rhizome extract of *D. cochleata* (D. Don) C

Sr. number	Name of the compound	Chemical structure of the compound	Compound nature
1	Dimethyl sulfoxide		Colorless liquid
2	Neoisolongifolene, 8,9-dehydro		Solid
3	Germacrene D		Volatile liquid
4	Cyclopropane		Liquid

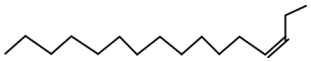
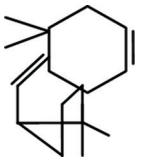
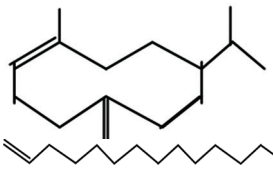
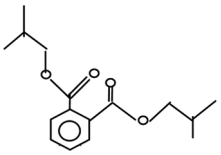
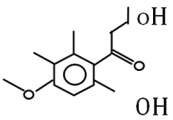
GC-MS: Gas chromatography-mass spectroscopy, *D. cochleata: Dryopteris cochleata*

Table 3: GC-MS analysis revealed the presence of phytochemical components in chloroform rhizome extract of *D. cochleata* (D. Don) C

Peak number	Retention time	Peak area (%)	Name of the compound	Molecular weight	Molecular formula
1	16.825	1.05	3-hexadecene, (Z)	224	$C_{16}H_{22}$
2	17.933	3.30	1,3-cyclohexanedione, 2-methyl-2-(3-oxobutyl)	196	$C_{11}H_{16}O_3$
3	18.250	2.34	Neoisolongifolene, 8,9-dehydro	202	$C_{15}H_{22}$
4	18.483	8.14	Germacrene D	204	$C_{15}H_{24}$
5	18.717	5.16	Phenol, 3,5-bis (1,1-dimethylethyl)	206	$C_{14}H_{22}O$
6	19.150	7.43	Cyclohexane, 1,1,3-trimethyl-2-(3-methylpentyl)	210	$C_{15}H_{30}$
7	19.425	1.45	1,6,10-dodecatrien-3-ol, 3,7,11-trimethyl	222	$C_{15}H_{26}O$
8	19.650	2.58	1-Pentadecene	210	$C_{15}H_{30}$
9	19.733	0.70	Hexadecane	226	$C_{16}H_{34}$
10	20.200	7.43	Pentyl filicinate	224	$C_{13}H_{20}O_3$
11	22.133	2.88	1-heptadecene	238	$C_{17}H_{34}$
12	23.242	4.18	1,2-benzenedicarboxylic acid, bis (2-ethylpropyl) ester	278	$C_{16}H_{22}O_4$
13	24.158	4.95	Aspidinol	224	$C_{12}H_{16}O_4$
14	26.758	1.40	1-nonadecene	266	$C_{19}H_{38}$

D. cochleata: Dryopteris cochleata

Table 4: Chemical structure of the most prevailing compounds of chloroform rhizome extract of *D. cochleata* (D. Don) C

Sr. number	Name of the compound	Chemical structure of the compound	Compound nature
1	3-hexadecene, (Z)		Solid
2	Neoisolongifolene, 8,9-dehydro		Solid
3	Germacrene D		Volatile liquid
4	1-pentadecene		Solid
5	Hexadecane		Liquid
6	1-heptadecene		Solid
7	1,2-benzenedicarboxylic acid, bis (2-methyl propyl) ester		Solid
8	Aspidinol		Solid
9	1-nonadecene		Liquid

D. cochleata: Dryopteris cochleata

ester ($C_{16}H_{22}O_4$) with retention time 23.242 and peak area of 4.18, Aspidinol ($C_{12}H_{16}O_4$) with retention time 24.158 and peak area of 4.95, 1-nonadecene ($C_{19}H_{38}$) with retention time 26.758 and peak area of 1.40. The other less prominent peaks at other retention times are showing the peak identities of the various compounds identified as shown in Fig. 2 and Table 3. The structure of prevailing compounds of chloroform rhizome extract is presented in Table 4.

The GC-MS analysis of *D. cochleata* rhizome in ethanol extract shows various types of compounds *viz*., dimethyl sulfoxide, Germacrene D, neoisolongifolene, 8, 9-dehydro and cyclopropane. The dimethyl sulfoxide has been used against cancer. It was used in pharmaceutical as an anti-inflammatory and antioxidant [6]. Germacrene D has anti-inflammatory properties [4]. Cyclopropane is used in clinical administration. Similarly, GC-MS analysis of chloroform extract shows various types [9] of compounds *viz*., 3-hexadecene, neoisolongifolene, 8,9-dehydro, Germacrene D, 1-pentadecene, hexadecane, 1-heptadecene, 1,2-benzenedicarboxylic acid, bis (2-methyl propyl) ester, aspidinol, and 1-nonadecene. Among these, 3-hexadecene was present which shows highly medicinal properties to cure Diabetes, inflammatory diseases, and cancers [12]. However, other compounds *viz*., 1-heptadecene, 1-nonadecene, hexadecane, 1-pentadecene were commonly used in many different fields of medicine. The compound aspidinol has anthelmintic, alterative astringent properties. The identifying of above compounds may be valuable in food and pharmaceutical industries [7,8].

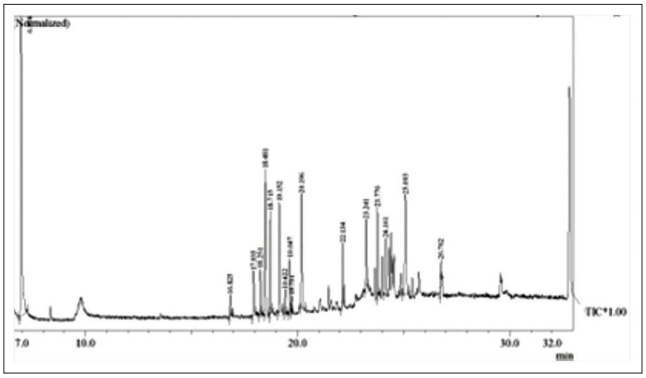


Fig. 2: GC-MS chromatogram of chloroform extract of whole plant extract of *Dryopteris cochleata* (D. Don) C

CONCLUSION

The aim of the present study was to provide information about the essential phytochemical constituents of *D. cochleata* (D. Don) C. rhizome. The result from the present investigation is very encouraging and indicates that this plant may be used as plant medicinal nutritive. The common three compounds observed in these two extracts are Germacrene D, 1, 3-cyclohexanedione, 2-methyl-2 (3-oxobutyl), neoisolongifolene, 8, 9-dehydro shows important role in the medicinal properties of pteridophyte.

Therefore, present investigation emphasizes the efficacy of traditional remedies and that it inspires the people to realize the importance of natural resources for its potent pharmaceutical use. It also provided as the basis for assessment of the preventive role of *D. cochleata* rhizome against free radical effects.

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