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Riya K
MPharm Final Year Student, Department of Pharmacognosy and Phytochemistry Nehru College of Pharmacy Pampady, Thiruvilwamala, Thrissur, Kerala, India

Dr. B Sreedharren
Principal, Department of Pharmacognosy and Phytochemistry Nehru College of Pharmacy Pampady, Thiruvilwamala, Thrissur, Kerala, India

Corresponding Author:
Dr. B Sreedharren
Principal, Department of Pharmacognosy and Phytochemistry Nehru College of Pharmacy Pampady, Thiruvilwamala, Thrissur, Kerala, India

GC-MS analysis of volatile phytocomponents from ethanolic plant extract of *Selaginella involvens* (Sw.) Spring

Riya K and Dr. B Sreedharren

Abstract
This investigation was carried out to determine the possible volatile phytocomponents present in the plant of *Selaginella involvens* Spring by using GC-MS analysis. The dried and pulverised plant material were extracted with ethanol (70%) for 6 hours. Our results of GC-MS compounds in the extract was relevant to NIST 11 library and WILEY 8. Total 12 compounds were identified. The volatile components in *Selaginella involvens* plant were made up largely of sesquiterpenoids, including neophytadiene (38.03%), followed by 3,7,11,15-Tetramethyl-2-hexadecen-1-ol (26.74%), (+-) lavandulol acetate (7.43%), 1-ethoxy-propene (5.81%), decanoic acid (5.29%), methyl 2-[(aminosulfonyl)methyl] benzoate (4.12%), 6-octen-1-ol, 3,7-dimethyl-,propanoate (3.79%) were found as the major compound. Small quantity of 2, 7-octadieniol acetate (2.49%), Hexane, 2,3-dimethyl-(2.46%), 7-dodecen-1-ol,(z)-(2.13 %), 3,5-dimethyl-4-deuteroxymethyl-isoxazole (1.26%), 1-methylbutyl nitrite (0.45 %) were present. This study result will make a way for the production of herbal medicines for various ailments by using *Selaginella involvens* Spring.

Keywords: 70% Ethanol, sesquiterpenoid, *Selaginella involvens* (Sw.) Spring, Western Ghats

Introduction
Nature has been a source of medicinal drugs for thousands of years.A lot of modern drugs have been isolated from natural sources. Many of these isolations were based on its traditional uses. This plant-based, medicine plays an essential role in health care. The genus Selaginella belonging to the family Selaginellaceae is composed of about 700 species. Although many ferns, are used in ethno-medical practices in remote villages in India, most of them are not pharmacologically evaluated. Among pteridophytes, Selaginella [Family: Selaginellaceae] is an important, unexplored genus with regard to their medicinal values. Some of the species of this genus are used in folk medicine as a guarded secret. *S. involvens, S. delicatula*. and *S. wightii* are three important Selaginella species found in the Western Ghats of Kerala. *S. involvens* is an epiphytic fem which also grows as lithophytes and terrestrial plants. It is said to possess the property of prolonging life and ameliorating old age related health problems including infections. *S. delicatula* is used in ethnomedicine for healing external ulcers and it is found throughout the southern parts of Western Ghats partially in shaded and moist areas. *S. wightii* is a rare prostrate fern found in open rocky areas. It is used in folk medicine to treat urinary infections. Selaginella plant species are used to reduce high fever and also considered as a strong anti- poison. *Selaginella involvens* has been used as complementary and alternative medicines in several traditional medication. It is traditionally used to cure wound, after childbirth, menstrual disorder, skin disease, headache, fever, infection of exhalation channel, infection of urethra, cirrhosis, cancer, rheumatism, bone fracture, etc. Part to be used is entire plant.

Selaginella species contains large number of bioactive compounds, which includes alkaloid, phenols, sterol, flavonoids, and terpenoid. Biflavonoids, such as amentoflavone, sumaflavone, robustaflavone, ginkgetin, hinokiflavone and isocryptomerin, are the most important valuable natural products of Selaginella and show various pharmacological activities including antioxidant, anti-inflammatory and antitumor. The medicinal use of Selaginella is occurred in the entire world. The largest usage is conducted by Chinese, especially for *S. tamariscina*, *S. doederleinii*, *S. moellendorffii*, *S. Uncinata*. Being an important ethnomedicinal herb, not much studies has been carried out on indian species of *Selaginella involvens* (Sw.) Spring. With this note the present study was designed to investigate GC-MS analysis of volatile phytocomponents from ethanolic extract of whole plant of *Selaginella involvens* Spring.

Materials and Methods

1. Plant collection and Authentication

The whole plant of *Selaginella involvens* Spring (Selaginellaceae) were collected from Rayirath Garden, Pattikkad, Thrissur district, Kerala, India. The collected materials were taxonomically identified by Dr.P.S.UDAYAN, Assistant professor Department of Botany and Research centre, Sreekrishna College, Guruvayur, kerala. The voucher specimen of *Selaginella involvens* (Sw.) Spring (84) was submitted in the Herbarium.

2. Preparation of Extract

The shade dried coarsely powdered whole plant of *Selaginella involvens* Spring was extracted with 70% ethanol by using soxhlet apparatus for 6 hours.The extract was filtered through Whatman filter paper No.1 and concentrated in vaccum rotary evaporator. This ethanolic extract was used for GC-MS analysis.

3. GC-MS analysis

Gas chromatography Mass spectrometry analysis of ethanolic extract was performed using Shimadzu GC-MS Model number: QP2010S equipped with Column - ELITE-5MS (30 meter length, 0.25 mm ID, and 0.25 µm thicknesses). Electron ionization system was used; details of GC programme were given in Table I. The oven temperature was programmed from 70.000C which is given in Table II. Helium gas was used as the carrier gas. Details of GC-MS programme was given in Table III. Programme specifications regarding Mass Spectra were depicted in Table IV.

GCMS Software: GCMS Solutions
Libraries used: NIST 11& WILEY 8.

Table 1: GC programme (GC 2010)

Column temperature	70.0^{0}C
Injection temperature	260.00^{0}C
Injection mode	Split
Sampling time	2.00 min
Flow control mode	Linear velocity
Pressure	61.5 pka
Total flow	54.1mL/min
Column flow	1.00mL/min
Linear velocity	36.7 cm/sec
Purge flow	3.0mL/min
Split ratio	50.0

Table 2: Oven temperature programme

Rate	Temperature(^{0}C)	Hold time(min)
-	70.0	2.00
10.00	200.0	5.00
5.00	280.0	15.00

Table 3: GC MS programme (GCMS-QP2010)

Ion source temperature	200.00^{0}c
Interface temperature	280.00^{0}c
Solvent cut time	6.50 min
Detector gain mode	Relative
Detector gain	1.01 kV+0.00 kV
Threshold	1000

Table 4: MS table

Start time	6.70 min
End time	51.00 min
ACQ time	Scan
Event time	0.50 sec
Scan speed	1000
Start m/z	50.00
End m/z	500.00
Sample inlet unit	GC

Results and Discussion

The ethanolic extract of *Selaginella involvens* (Sw.) Spring was carried out by soxhlet extraction method and the volatile phytocomponents of the extract was analysed using GC-MS. Totally 12 compounds were detected through GC-MS, and depicted in Table 1. All the twelve compounds were identified by comparison with the authentic spectra obtained from GC-MS library (NIST - 11 and WILEY 8) with SI factor. Of the twelve compounds identified from ethanolic extract Neophytadiene (38.03%) was found as a major compound which is an sesquiterpenoid. The second major compound was detected as 3,7,11,15-Tetramethyl-2-hexadecen-1-ol (Phytol) (26.74%) is an acyclic diterpene alcohol. The third major chemical constituent was (+-) - Lavandulol acetate which comes under aliphatic monoterpenoid alkene ester which occupied 7.43% of the total extract. Neophytadiene have antipyretic, analgesic, anti-oxidant, anti inflammatory, antimicrobial properties, also used in the treatment of headache, rheumatism, some skin disease. Phytol possess antimicrobial, anticancer, antiinflammatory, anti diabetic, anti diuretic, and immunostimulatory activities. Lavandulol acetate is used for supporting respiratory health, improves appearance of skin and hair, relieves pain and also used as a perfuming agent. The other compounds *viz*. 1-ethoxy-propene (5.81%), decanoic acid (5.29%), methyl 2-[(aminosulfonyl)methyl] benzoate (4.12%), 6-octen-1-ol, 3,7-dimethyl-,propanoate (3.79%) 2,7-octadieniol acetate (2.49%), Hexane, 2,3-dimethyl- (2.46%),7-dodecen-1-ol,(z)- (2.13%),3,5-dimethyl- 4-deuteroxymethyl -isoxazole (1.26%), 1-methylbutyl nitrite (0.45%) were found as a meagre amount in the extract. The plant has the promising bioactive compounds which are helpful in the production of novel pharmaceuticals.

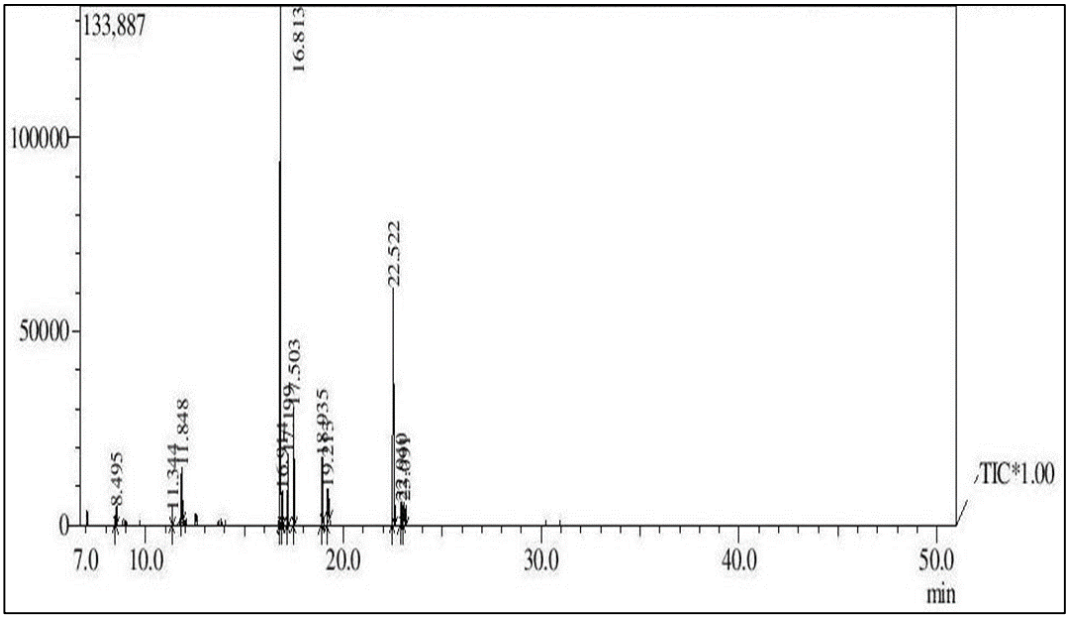

Fig 1: GC-MS chromatogram of ethanolic extract of *Selaginella involvens* (Sw.) Spring

Table 5: GC-MS analysis of volatile phytocomponents from ethanolic extract of *Selaginella involvens* (Sw.) Spring

Peak#	Rt	Area	Area%	Height	Height %	Name	Base m/z
1	8.495	10339	1.26	4588	1.47	3,5-Dimethyl-4-Deuteroxymethyl-isoxazole	128.05
2	11.344	3734	0.45	3344	1.07	1-Methyl butyl nitrite	57.05
3	11.848	47731	5.81	13212	4.25	1- Ethoxy-propene	57.05
4	16.813	312465	38.03	133887	43.03	Neophytadiene	68.05
5	16.914	20203	2.46	8984	2.89	Hexane, 2,3-dimethyl-	70.05
6	17.199	31123	3.79	18280	5.88	6- Octen-1-ol,3,7-dimethyl-,propanoate	82.10
7	17.503	61013	7.43	30426	9.78	(+-)-Lavandulol acetate	82.10
8	18.935	43469	5.29	17047	5.48	Decanoic acid	73.05
9	19.213	33865	4.12	8809	2.83	Methyl 2-[(aminosulfonyl)methyl]benzoate	149.05
10	22.522	219754	26.74	60938	19.59	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	57.05
11	22.940	17528	2.13	5843	1.88	7-Dodecen-1-ol,(z)-	67.05
12	23.091	20448	2.49	5775	1.86	2,7-Octadieniol acetate	67.05
		821672	100.00	311133	100.00		

Conclusion

In the present investigation twelve volatile phytocomponents have been identified from ethanolic plant extract of *Selaginella involvens* (Sw.) by Gas Chromatography Mass Spectrometry (GC-MS) analysis. The presence of various phytochemicals contributes to the various bioactive compounds have different medicinal properties which can be useful for the treatment of several diseases.

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Conflit of Interest

No conflict of interest

Abbreviation Used

GC-MS: Gas Chromatography – mass spectrometry; Rt: Retention time; RI: Retention index; SI: Super Impossibility

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