

GC-MS Analysis of *Bulbophyllum Kaitense* Rechib. Pseudobulbs Eastern Ghats of India

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Abstract

The active constituents present in Pseudobulbs parts of *Bulbophyllum kaitense* Rechin. (Orchidaceae). Twenty Five compounds in Ethanol extract were identified Bioactive by Gas chromatography –Mass spectrometry (GC-MS). This analysis revealed that *Bulbophyllum Kaitense* Pseudobulbs contain mainly 1,14 – Tetradecanediol (4.023%) 10- Undecyn -1-01 (10.74%) 1, Octanol, 2 – butyl – (7.20%) 2H-1- Benzopyran -6 -01, 3, 4 - dihydro – 2,8 – dimethyl -2 – (4,8,12 – trimethyl tridecyl) –[2R –[2R*(4R*,8R*)]] – [e’ – Tocopherol]. Squalene (1.53%) 3,3’, 4,4’ - Tetramethoxystilbene (1.17%) 2-(2- Hydroxy – 2 – p - methoxyphethyl) - 5- methyl (1.09%) Ethyl homovanillate (0.97%) 2 (1H) – Phenanthrene, 3,4,4a, 9,10,10a- Hexahydro – b- hydroxyl -1,1, 4a- trimethyl -7- (1- methylethyl.) (4aS-trans) – (0.80%) Decanoic acid. Ethyl ester (0.76%). This is the first report of identification of active constituent from Pseudobulbs parts of *Bulbophyllum Kaitense* by GC-MS.

Keywords: *Bulbophyllum Kaitense*, Constituents, GC-MS Analysis.

Introduction

Most traditional Medicines are developed from nature. They have not yet fulfilled the scientific requirements so as to be classified as modern medicines. For purpose of scientific back up, a study is need to examine their bioactive components, their efficacy and safety, usually, most components that are useful for medicinal purposes are secondary metabolites. In the development of medicinal plant industry, plant medicines are classified into Three group’s herbs (Jammu.) Standardized extracts and phyto pharmaceuticals. There are strict requirements for standardizing the extracts.

Some of them include correctness and proven restorative power, uniformity of active constituents, their efficacy, safety and assurance, both in quality and quantity.

The endemic plants of South India. Orchidaceae is one of the largest families among angiosperms. According to one estimate the family includes 800 genera and 25,000 species. Orchids are well known for their economic importance and widely cultivated for ornamental purposes. Orchids are cosmopolitan in distribution. *Vanilla planifolia* is a commercially important orchid as it is the source of vanilla used as a foodstuff flavoring. This may account for the use of orchids as aphrodisiacs in ancient civilizations. When we study the history of ancient alternative systems of medicine Ayurveda and Traditional Chinese Medicine (TCM) are on the forefront.

Bulbophyllum kaitense is an epiphytic family orchidaceous. Endemic to south India. The plant is dense mats on trees and rock. It is native of India. Occurs in the forest of Eastern Ghats from Kollihills above 1300m. Sympodial epiphytes with pseudobulbs. On the rhizome. Terminating the pseudobulbs. Inflorescence umbellate scape. Pseudobulb's greenish. Subfusiform not angled 2cm long 4-5 cm part on the zone. Leaves 9-13cm long flowers without mentum. Sepal unequal petals shorter than lateral sepals. The plants have been used in indigenous medicine such as Ayurveda and local traditional medicinal practices the pseudobulb is used for the treatment of certain antioxidant, anticancer, anti-inflammatory, antiseptic, antitumor and antimicrobial activity, the pseudobulb property in curing of different diseases. This part was selected for the study. Hence the present investigation was carried out to determine the possible chemical components from *Bulbophyllum kaitense* pseudobulb.

Materials and Methods

Plant Material

The plant material *Bulbophyllum kaitense*. Plants were collected from Eastern Ghats of Kollihills in Namakkal District of Tamil Nadu, India. The botanical identification of the plant was confirmed by Ret, Dr. S. John Britto, The Director, The Rapinat Herbarium and centre for molecular systematic, St. Joseph's college (Campus) Tiruchirappalli-620020. Tamil Nadu, India. The plant voucher number: RHT.872.

Plant Sample Extraction

20 gm powdered plant material is soaked in 50 ml of absolute ethanol overnight and then filtered through Whatman filter paper No.41 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 is wetted with ethanol the filtrate is then concentrated by bubbling nitrogen gas into the solution and reduces the volume to 1ml the extract contains both polar and non-polar phytochemicals.

GC-MS Analysis

GC-MS analysis was carried out on a GC Clarus 500 Perkin Elmer system comprising a AOC-20i autosampler and gas chromatograph interfaced to a mass spectrometer.

(GC-MS) instrument employing the following conditions. Column Elite-5MS fused Silica capillary column (30mmX0.25mmX0.25 μ mdf, composed of 5% Diphenyl / 95% Dimethyl polySiloxane), operating in electron impact mode at 70eV; Helium(99.999%) was used as carrier gas at a constant flow of 0.1ml min and an injection volume of 2 μ l was employed (split ratio of 10:1) injector temperature 250 $^{\circ}$ C; Ion- source temperature 280 $^{\circ}$ C. The oven temperature was programmed from 110 $^{\circ}$ C (isothermal for 2min), with an increase of 10 $^{\circ}$ C/min, to 200 $^{\circ}$ C, then 5 $^{\circ}$ C/min to 280 $^{\circ}$ C, ending with a 9min isothermal at 280 $^{\circ}$ C mass spectra were taken at 70eV; a scan interval of 0.2 seconds and fragments from 40 to 450 Da. Total GC running time is 36min

Identification of Components

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 components was compared with the spectrum of the known compounds stored in the NIST library. The name, molecular weight and structure of the components of the test materials were ascertained.

Results and Discussions

Twenty five bioactive compounds were identified in *Bulbophyllum Kaitense* Rechib. Pseudobulb by GC-MS analysis. The active principles with their retention time (RT), molecular formula. Molecular weight (MW) and concentration (%) are presented in (Table 1 and fig 1) 1,14 – tetradecanediol (40.23%) 10- Undecyn -1-01(10.74%) 1-Octano, 2-butyl – (7.20%). 2H-1- Benzopyran -6 -01, 3,4 – dihydro -2, 8 – dimethyl – 2 – (4,8,12 - trimethyltridecyl) – [2R-[2R*(4R*,8R*)]] – [-e” - Tocopherol]. Squalene (1.53%) 3,3',4,4' - Tetramethoxystilbene (1.17%) 2- (2- Hydroxy – 2 – p - methoxyphenethyl) - 5- methyl (1.09%) Ethyl homovanillate (0.97%) 2 (1H) – Phenanthrene, 3,4,4a, 9,10,10a- Hexahydro – b- hydroxyl -1,1, 4a- trimethyl -7- (1-methylethyl.) (4aS-trans) – (0.80%) Decanoic acid. Ethyl ester (0.76%).

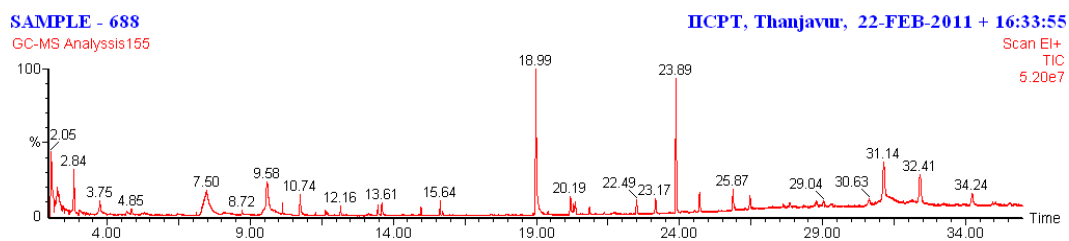


Figure 1: GC-MS Chromatogram of ethanolic extract of *Bulbophyllum kaitense*. Pseudobulb.

Table 1: Activity of Phyto-Components identified in the ethanolic extract of *Bullbophyllum kaitense*. Pseudobulb by GC-MS.

No	RT	Name of the compound	Molecular Formula	M W	Peak Area %	Nature of compound	**Activity
1.	2.26	Butane, 1,1-diethoxy-3-methyl-	C ₉ H ₂₀ O ₂	160	3.10	Ether compound	No activity reported
2.	2.84	Propane, 1,1,3-triethoxy-	C ₉ H ₂₀ O ₃	176	1.45	Ether compound	No activity reported
3.	3.75	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	C ₆ H ₈ O ₄	144	0.52	Flavonoid fraction	Antimicrobial Antiinflammatory
4.	4.85	Butanoic acid, 3-oxo-, hexyl ester	C ₁₀ H ₁₈ O ₃	186	8.41	Ester compound	No activity reported
5.	7.50	Sucrose	C ₁₂ H ₂₂ O ₁₁	342	5.71	Sugar compound	Preservative
6.	9.58	1,2,3,5-Cyclohexanetetrol, (1à,2á,3à,5á)-	C ₆ H ₁₂ O ₄	148	0.32	Poly Hydroxy compound	Analgesic, Anesthetic, Antioxidant, Antiseptic, Antibacterial, Antiviral Cancer preventive
7.	10.14	à-Bisabolol	C ₁₅ H ₂₆ O	222	1.29	Sesquiterpene alcohol	Antimicrobial Anticancer Antiinflammatory
8.	10.74	4-((1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol	C ₁₀ H ₁₂ O ₃	180	0.32	Phenolic compound	Analgesic, Anesthetic, Antioxidant, Antiseptic, Antibacterial, Antiviral Cancer preventive, Fungicide, Rodenticide Emetic, Vasodilator.
9.	11.63	1,14-Tetradecanediol	C ₁₄ H ₃₀ O ₂	230	40.23	Alcoholic compound	Antimicrobial
10.	12.16	1-Heptadecanol	C ₁₇ H ₃₆ O	256	0.40	Saturated alcoholic compound	Antimicrobial
11.	13.47	Decanoic acid, ethyl ester	C ₁₂ H ₂₄ O ₂	200	0.76	Lauric acid ester	Antioxidant, Antibacterial, COX-1 & COX-2 inhibitor, Antiviral, Hypocholesterolemic, Candidicide.
12.	13.61	2-Adamantanol, 6,6-ethylenedioxy-	C ₁₂ H ₁₈ O ₃	210	0.36	Alcoholic compound	Antimicrobial
13.	14.98	Phytol	C ₂₀ H ₄₀ O	296	0.64	Diterpene	Antimicrobial Anticancer Antiinflammatory Diuretic
14.	15.64	13-Tetradecene-11-yn-1-ol	C ₁₄ H ₂₄ O	208	0.20	Alcoholic compound	Antimicrobial

15.	15.7 3	10-Undecyn-1-ol	C ₁₁ H ₂₀ O	168	10.7 4	Alcoholic compound	Antimicrobial
16.	18.9 9	2-(2-Hydroxy-2-p-methoxyphenylethyl)-5-methyl	C ₁₄ H ₁₆ N ₂ O ₂	244	1.09	Nitrogen compound	Antimicrobial
17.	20.1 9	Ethyl homovanillate	C ₁₁ H ₁₄ O ₄	210	0.97	Phenolic compound	Antimicrobial Antioxidant
18.	22.4 9	S-Indacene-1,7-dione, 2,3,5,6-tetrahydro-3,3,4,5,5,8-hexamethyl-	C ₁₈ H ₂₂ O ₂	270	0.97	Ketone compound	No activity reported
19.	23.1 7	1-Octanol, 2-butyl-	C ₁₂ H ₂₆ O	186	7.20	Alcoholic compound	Antimicrobial;
20.	23.8 9	3,3',4,4'-Tetramethoxystilbene	C ₁₈ H ₂₀ O ₄	300	1.17	Stilbene compound	Anticancer
21.	24.7 1	Squalene	C ₃₀ H ₅₀	410	1.53	Triterpene	Antibacterial, Antioxidant, Antitumor, Cancer preventive, Immunostimulant, Chemo preventive, Lipoxygenase- inhibitor, Pesticide
22.	25.8 7	2(1H)-Phenanthrenone, 3,4,4a,9,10,10a-hexahydro-6-hydroxy-1,1,4a-trimethyl-7-(1-methylethyl)-, (4aS-trans)-	C ₂₀ H ₂₈ O ₂	300	0.80	Phenolic compound	Antimicrobial Antioxidant
23.	26.4 7	2H-1-Benzopyran-6-ol, 3,4-dihydro-2,8-dimethyl-2-(4,8,12-trimethyltridecyl)-, [2R-[2R*(4R*,8R*)]]-[.ë-Tocopherol]	C ₂₇ H ₄₆ O ₂	402	7.08	Vitamin E	Antiageing, Analgesic, Antidiabetic Antiinflammatory, Antioxidant, Antidermatitic, Antileukemic, Antitumor, Anticancer, Hepatoprotective, Hypocholesterolemic, Antiulcerogenic, Vasodilator, Antispasmodic, Antibronchitic, Anticoronary
24.	31.1 4	Stigmasterol	C ₂₉ H ₄₈ O	412	3.90	Steroid	Antimicrobial Diuretic Anti-inflammatory Antiasthma Antiarthritic Anticancer
25.	32.4 1	2H-Pyran, 2-(7-heptadecyloxy)tetrahydro-	C ₂₂ H ₄₀ O ₂	336	3.10	Flavonoid fraction	Antimicrobial Antiinflammatory

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