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Volatile constituents of *Nardostachys jatamansi* DC., a critically endangered species

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ABSTRACT

The hydro-distilled volatile oil of *Nardostachys jatamansi* rhizomes was analyzed using GC and GC-MS. The volatile oil was consists of large number of sesquiterpenes (76.65 %) and aliphatic components (16.29 %) while monoterpenes (5.11 %) and diterpenes (1.48 %) were present in less amounts. Prominent sesquiterpenes were *t*-cadinol (22.67 %), *α*-eudesmol (3.00 %), 5-*neo*-cedranol (2.51 %), muurolol (1.37 %) and jatamansone or valeranone (36.71 %). Among fourteen aliphatic components, there were six aliphatic hydrocarbons (1.47 %), three were aliphatic alcohols (0.47 %), one aliphatic aldehyde (0.04 %) and two each aliphatic esters (12.74 %), and aliphatic acids (1.57 %). Hexadecanoic ethyl ester and tetradecanoic acid (1.51 %) were the predominent among aliphatic components.

KEYWORDS

Nardostachys jatamansi;
Volatile oil;
GC-MS;
Sesquiterpenes;
Jatamansone.

INTRODUCTION

Nardostachys jatamansi (D.Don) DC. (Family-Valerianaceae), a critically endangered rhizome-bearing medicinal plant, is restricted to specialized habitats in high altitudes of the Himalaya[1] from Pakistan, India (Jammu and Kashmir, Himachal Pradesh, Uttarakhand, Sikkim) to Nepal, Tibet and China between 3300 to 5000 m asl[2]. Due to overexploitation of rhizomes for medicinal and aromatic uses, habitat degradation and other biotic interferences, the species has been declared critically endangered and survival of the herbs is at risk[1-3].

Traditionally, jatamansi is used as tonic, stimulant and antiseptic and also used for the treatment of epilepsy, hysteria, convolutions, heart palpitation, intestinal colic and antiarrhythmic activities[4]. The plant has a rich history of medicinal use and has been valued for centuries in Ayurvedic (Indian) and Unani (ancient Greco-Arab) systems of medicine. In Ayurveda, roots and rhizomes of *N. jatamansi* are used to treat hysteria, epilepsy, and convulsions[5]. The decoction of the drug is also used in neurological disorders, insomnia and disorders of cardiovascular system[5-7]. In Unani System of Medicine, it is also known as *Sambul-ut-teeb* and widely used in many formulations for the treatment of various diseases *e.g. Safoof-e-Muhazzil* for obesity[8].

There are two species, *N. jatamansi* and *N. chinensis* widespread throughout the northern part of alpine to sub alpine Himalayan region. Rhizome is the source of Spikenard oil[9]. The principal constituents of *N. jatamansi* are essential oil (0.5-2%), rich in sesquiterpenes and coumarins[11]. Jatamansone or valeranone is the principal sesquiterpene[10,11]. Other

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sesquiterpenes include nardostachone, dihydrojatamansin, jatamansinol, jatamansic acid[12]. Various extracts and volatile oil of *N. jatamansi* was reported to exhibit antidepressant[13], antioxidant[14], GABA enhancing[15], cardio-protective and hypolipidemic[16], hepatoprotective[17], anticonvulsant[18], antiarthritic[19], antipyretic[20], antistress[21], antimicrobial, antifungal, insecticidal[22,23], gastrointestinal tract disorders[24], antiparkinson's[25], neuroprotective[26], tranquillizing activities[27]. The aim of this paper is to identify the chemical composition of the essential oil of *Nardostachys jatamansi* rhizomes by GLC and GC-MS analysis.

MATERIAL AND METHODS

Collection of plant material and authentication

The rhizomes of *Nardostachys jatamansi* were purchased from Samsi Dawakhana, Ballimaran, Delhi, a registered shop of Unani Medicine and authenticated by Dr. H. B. Singh, Scientist F and Head, Raw Materials Herbarium and Museum, National Institute of Science Communication and Information Resources (NISCAIR), New Delhi. Voucher specimen of drug was deposited in the Raw Materials Herbarium and Museum, National Institute of Science Communication and Information Resources (NISCAIR), New Delhi, with reference number Ref. NISCAIR/RHMD/consult/ -2010-11/1705/05.

Isolation of volatile oil

The drug (200 gm) was hydro-distilled for six hours with Clevenger apparatus. The yield of volatile oil obtained was 0.7 % v/w. The light green coloured volatile oil was collected in the graduated tube. The collected volatile oil was dried over anhydrous sodium sulphate and stored at 4 ^{0}C in the dark.

GC analysis

The gas chromatographic analysis of the volatile oil was carried out on Shimadzu 2010 Gas Chromatograph (Japan) equipped with a flame ionization detector (FID) and AB-Innowax 7031428 WCOT fused capillary column (60 m x 0.25 mm x 0.25 μm). The injector and detector (FID) temperatures were maintain at 250 and 270 ºC, respectively. The carrier gas used was nitrogen at a flow rate of 1.21 mL/min with column pressure of 155.1 kPa. The sample (0.2 μl) was injected into the column with a split ratio of 80:1.

Component separation was achieved following a linear temperature programmed from 60-230 ºC at a rate of 3 ºC/min and then held at 230 ºC for 9 min, with a total run time of 55.14 min. Percentage of the constituents were calculated by electronic integration of FID peak areas.

GC-MS analysis

The analysis of the volatile constituents were run on a Shimadzu QP-2010 GC-MS system equipped with AB-Innowax 7031428 WCOT column (60 m x 0.25 mm x 0.25 μm) directly coupled to the MS. The carrier gas was helium with a flow rate of 1.21 mL/min. oven temperature was programmed as 50 ºC for 1 min and subsequently held isothermal for 2 min. injector port: 250 ºC, detector: 280 ºC, split ratio 1:50, volume injected: 1 μL of the oil. The recording was performed at 70 eV, scan time 1.5 s; mass range 40-750 amu. Software adopted to handle mass spectra and chromatograph was a Chem station (Figure 2).

Identification

The individual peaks/constituents were identified by gas chromatography by comparison of their retention indices (R.I.) either with those of authentic compounds available in author's laboratory or with those of literature in close agreement to R.I.[28-34]. Further identification was made by comparison of fragmentation pattern of mass spectra obtained by GC-MS analysis with those stored in the spectrometer database of NBS 54 K.L, WILEY8 libraries and published literature[20-26].Retention indices of the components were determined relative to the retention times of a series of n-alkanes relative to C_9-C_{20} on HPS and HP-20M columns.

RESULTS AND DISCUSSIONS

The volatile oil of *Nardostachys jatamansi* consists of large number of sesquiterpenes (76.65 %) and aliphatic components (16.29 %) while monoterpenes (5.11 %) and diterpenes (1.48 %) were present in fewer amounts as given in the previous reports (TABLE 1, Figure 1)[9-12].

Full Paper

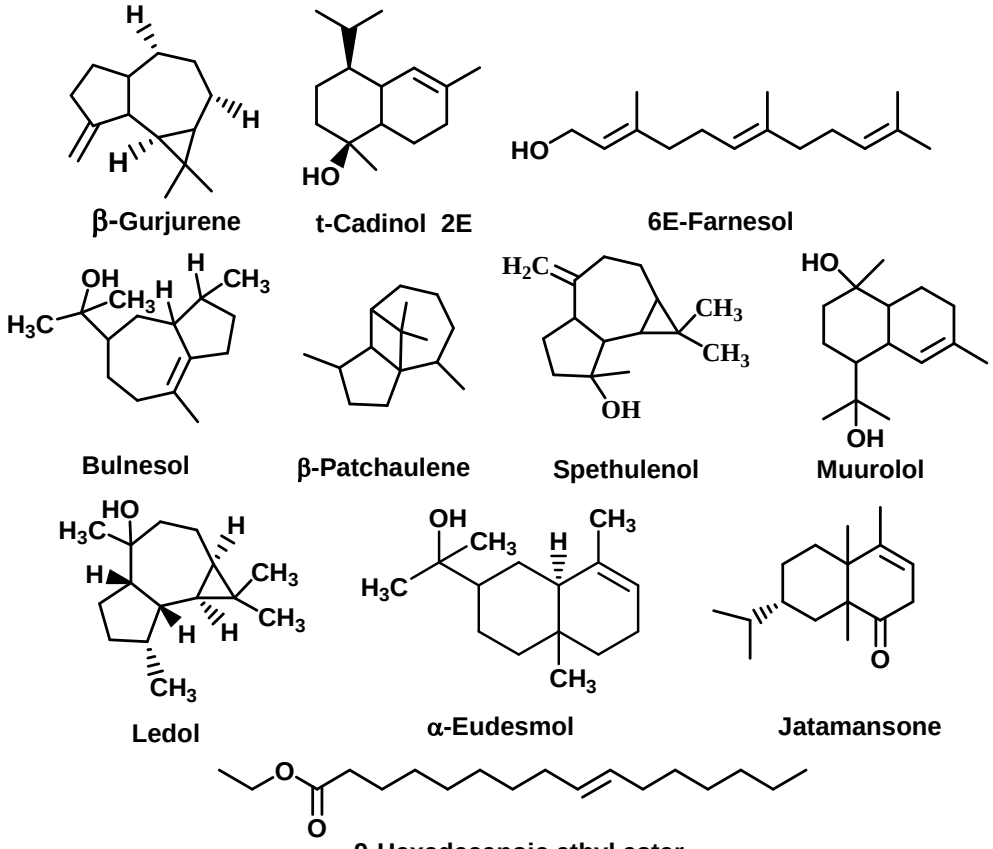

Figure 1 : Prominent components of volatile oil of *Nardostachys jatamansi* rhizome

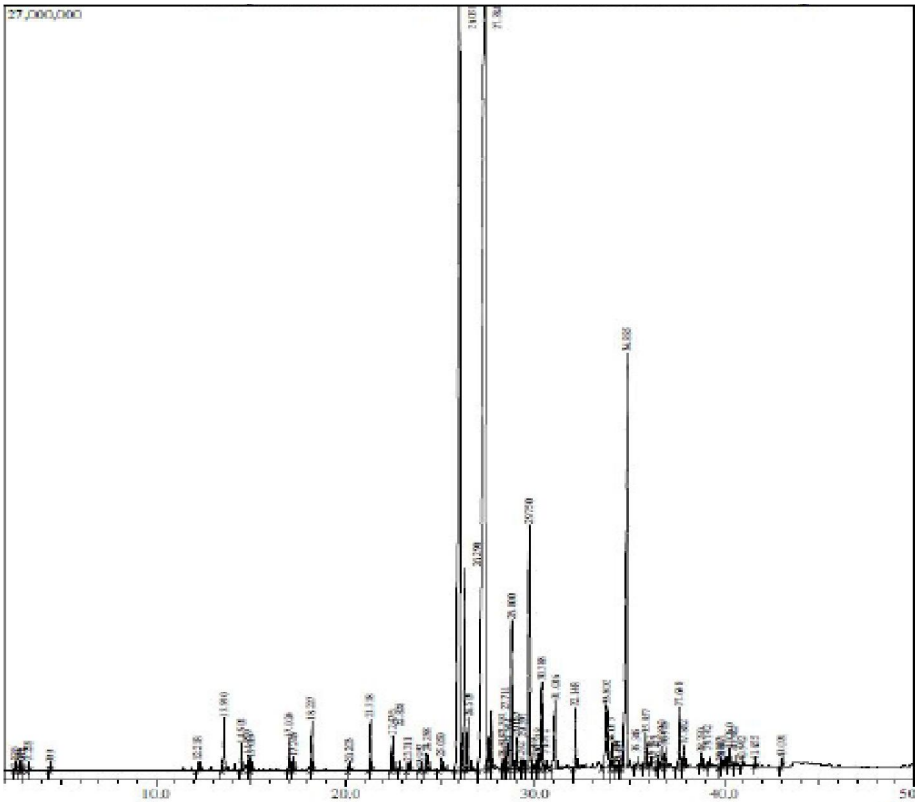

Figure 2 : GC-MS spectra of volatile oil of *Nardostachys jatamansi* rhizome

Full Paper

TABLE 1 : Volatile oil constituents of *Sambul-ut-Teeb* (*Nardostachys jatamansi DC.*)

S.No.	Components	Percent(%)	Kovats index	S.No.	Components	Percent(%)	Kovats index
1	α-Pinene	0.07	933	31	Eugenyl valerate	4.72	1718
2	β-Patchoulene	0.10	1378	32	Methyl heptadecane	0.11	1734
3	β-Gurjunene	0.68	1413	33	*n*-Pentadecanol	0.28	1776
4	γ-Elemene	0.31	1433	34	Tetradecanoic acid	1.51	1777
5	α-Humulene	0.20	1436	35	*n*-Octadecane	0.06	1795
6	Aromadendrene	0.14	1445	36	(2Z,6E)-Farnesyl acetate	1.76	1824
7	Alloaromadendrene	0.38	1465	37	Vomifoliol	1.10	1837
8	α-Selinene	0.16	1473	38	7-Hexadecenoic ethyl ester	1.49	1842
9	α-Panasinsen	0.56	1518	39	3-Methyl octadecane	0.51	1873
10	Nerolidol	0.08	1561	40	*n*-Nonadecane	0.05	1896
11	Ledol	0.59	1565	41	Hexadecanoic acid	0.06	1923
12	Spathulenol	0.42	1575	42	9-Hexadecenoic ethyl ester	11.25	1966
13	1-Caryophyllene oxide	0.13	1581	43	Menonyl oxide	0.04	1998
14	Globulol	0.09	1585	44	*n*-Eicosane	0.63	2001
15	Himachelene oxide	0.02	1610	45	Octadecanol	0.06	2080
16	Cubenol	0.20	1614	46	*n*-Heneicosane	0.11	2102
17	β-Eudesmol	0.17	1630	47	Manool	0.37	2105
18	t-Cadinol	22.67	1641	48	Unknown	0.35	-
19	α- Eudesmol	3.00	1649	49	Phytol	1.07	2011
20	Muurolol	1.37	1655	50	Unknown	0.34	-
21	Bulnesol	0.46	1664	51	Unknown	0.21	-
22	Jatamansone orValeranone	36.71	1667	52	Unknown	0.07	-
23	Epi-(E)-caryophyll-9-en-14-ol	0.83	1673	53	Unknown	0.07	-
24	*n*-Tetradecanol	0.13	1679	54	Unknown	0.09	-
25	α-Bisabolol	0.03	1685	55	Unknown	0.21	-
26	Eudesma-3,5-dien-1-ol	0.46	1691	56	Unknown	0.04	-
27	5-*neo*-Cedranol	2.51	1699	57	Unknown	0.04	-
28	*cis*-Farnesal	0.50	1705	58	Unknown	0.05	-
29	Hexadecanal	0.04	1712	59	Unknown	0.11	-
30	(2Z,6E)-Farnesol	0.20	1715				

Among three monoterpenes, two were monoterpene hydrocarbons (0.38 %), α-pinene and γ-elemene and one monoterpene ester, eugenyl valerate (4.72 %). Among twenty eight sesquiterpenes (76.65 %), seven were sesquiterpene hydrocarbons (2.22 %), sixteen were sesquiterpene alcohols (34.31 %), one sesquiterpene ketone (36.71 %), two oxides (0.15 %) and one each sesquiterpene ester (1.76 %) and sesquiterpene aldehyde (0.50 %). The sesquiterpene hydrocarbons were consists of α-panasinsen (0.56 %), alloaromadendrene (0.38 %), aromadendrene (0.14 %), α-selinene (0.16 %), β-gurjunene (0.68 %), α-humulene (0.20 %) and patchaulene (0.10 %). Among sixteen sesquiterpene alcohols, there were *t*-cadinol (22.67 %), α-eudesmol (3.00 %), 5-*neo*-cedranol (2.51 %), muurolol (1.37 %), vomifoliol (1.10 %), epi-(E)-caryophyllene-9-ene-14-ol (0.83 %), ledol (0.59 %), spethulenol (0.42 %), cubenol (0.2 %), β-eudesmol (0.17 %) and the other sesquiterpene alcohol were found in very less amount *e.g.* nerolidol (0.08 %), globulol (0.09 %), α-bisabolol (0.03 %) and (2Z,6E)-farnesol (0.2 %). Jatamansone or valeranone (36.71 %) was the predominant sesquiterpene ketone of the volatile oil[9]. Sesquiterpene oxides were present in less amount, caryophyllene oxide (0.13 %) and himachalene oxide (0.02 %). The

only one sesquiterpene ester and sesquiterpene aldehyde was (2Z,6E)-farnesyl acetate (1.76 %) and *cis*-farnesal (0.50 %), respectively[35].

Among fourteen aliphatic components (16.29 %), there were six aliphatic hydrocarbons (1.47 %), three aliphatic alcohols (0.47 %), one aliphatic aldehyde (0.04 %) and two each aliphatic esters (12.74 %), and aliphatic acids (1.57 %). 9-Hexadecanoic ethyl ester (11.25 %) and tetradecanoic acid (1.51 %) were the predominant among aliphatic components. The other aliphatic components were in fewer amounts *e.g.* hexadecanol (0.04 %), methyl heptadecane (0.11 %), n-pentadecanol (0.28 %), *n*-octadecane (0.06 %), 3-methyl octadecane (0.51 %), n-nonadecane (0.05 %), *n*-eicosane (0.63 %), octadecanol (0.06 %) and *n*-heneicosane (0.11 %). There were three diterpenes consisting of menoyl oxide (0.04 %), manool (0.37 %) and phytol (1.07 %). There were ten unknown (1.23 %) found in volatile oil.

Essential oil of its formulation "*Safoof-e-Muhazzil*" reported to contain many constituents *e.g.* eudes-4(14),11-diene (28.61 %), viridiflorol laurate (16.40 %), bisabolene (9.73 %), globulol (9.13 %), thymol (6.14 %), *t*-cadinol (4.15 %), *trans*-cadine-1,4-diene (2.08 %), 2E, 6E-farnesol (1.41), limonene (1.39 %), δ-cadinene (1.37 %) and β-gurjunene (1.28), which can be compared[35].

Essential oil of *N.jatamansi* from Kathmandu (Nepal) contains mainly β-patchoulene, β-gurjunene (29.10 %), δ-cadinene (0.98 %), γ-cadinene (0.81 %), cadinol (0.44 %), jatamansone (9.71 %), aristolenone (6.48 %)[10]. Essential oil from Lahore (Pakistan) reported to contain mainly ledene oxide [II] (13.021%), patchouli alcohol (9.582%), spathulenol (2.672%), globulol (1.876%), 4-[3,3-dimethyl-but-1-ynyl]-4-hydroxy-2,6,6-trimethylcyclohex-2-enone (1.849%), magastigma-4,6[E], 8[Z]-triene (1.015%), aristolene (0.997%) and β -vatirenene (0.932%)[36].

Variation in the composition of essential oils depends on their geography, time of collection, stages of plant growth and seasonal and environmental factors. Variations in the traded herbal composition occurs on account of geo-climatic conditions of their growth, maturity at the time of collection, species variation at times, substitutability on the basis of perceived efficacy and dubious trade practices[37].

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Full Paper

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