

GC-MS Analysis of Volatile Organic Constituents of Traditionally Used Medicinal Plants from the Western Ghats of India: *Blumea lanceolaria* (Roxb.) Druce., *Heliotropium indicum* L. and *Triumfetta rhomboidea* Jacq.

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Received September 9th, 2019; Accepted January 8th, 2020.

DOI: <http://dx.doi.org/10.29356/jmcs.v64i2.1093>

Abstract. The present study explored essential oil constituents of the medicinal plants used in traditional medicine and growing wild in the Western Ghats region of India, which is one of the 34 global biodiversity hotspots. The hydro distilled essential oils of *Blumea lanceolaria* (Roxb.) Druce. (Asteraceae), *Heliotropium indicum* L. (Boraginaceae) and *Triumfetta rhomboidea* Jacq. (Tiliaceae) were analyzed using gas chromatography equipped with a flame ionization detector (GC-FID) and gas chromatography coupled with mass spectroscopy (GC/MS). Thirty-six, twenty-seven, and fifty constituents were identified from the essential oils of *B. lanceolaria*, *H. indicum*, and *T. rhomboidea*, respectively comprising 97.3% (*B. lanceolaria*), 98.2% (*H. indicum*) and 97.6% (*T. rhomboidea*) of the total oil constituents. The major compounds of *B. lanceolaria*, *H. indicum*, and *T. rhomboidea* were identified as β -pinene (82.3%), methyl salicylate (54.3%), and β -caryophyllene (28.9%), respectively. The essential of *B. lanceolaria* was found to be rich in monoterpene hydrocarbons (90.4%), while *H. indicum* and *T. rhomboidea* oils were rich in phenyl derivative (72.8%) and sesquiterpene hydrocarbons (69.5%) type constituents, respectively. The common compounds viz., terpin-4-ol and eugenol were identified in the essential oils of *B. lanceolaria*, *H. indicum*, and *T. rhomboidea*.

Keywords: *Blumea lanceolaria* (Roxb.) Druce.; *Heliotropium indicum* L.; *Triumfetta rhomboidea* Jacq.; terpin-4-ol; eugenol; GC/MS.

Resumen. El presente estudio exploró los componentes de los aceites esenciales de plantas medicinales utilizadas en la medicina tradicional y de plantas silvestres que crecen en la región occidental de Ghats, India, que es uno de los 34 puntos críticos de la biodiversidad mundial. Los aceites esenciales hidro destilados de *Blumea lanceolaria* (Roxb.) Druce. (Asteraceae), *Heliotropium indicum* L. (Boraginaceae) y *Triumfetta rhomboidea* Jacq. (Tiliaceae) se analizaron mediante cromatografía de gases equipada con un detector de ionización de flama (GC-FID) y por cromatografía de gases acoplada con espectrometría de masas (GC / MS). Se identificaron treinta y seis, veintisiete y cincuenta componentes de los aceites esenciales de *B. lanceolaria*, *H. indicum* y *T. rhomboidea*, respectivamente, que comprenden 97.3% (*B. lanceolaria*), 98.2% (*H. indicum*) y 97.6 % (*T. rhomboidea*) del total de los constituyentes del aceite esencial. Los principales compuestos de *B. lanceolaria*, *H. indicum* y *T. rhomboidea* se identificaron como β -pineno (82.3%), salicilato de metilo (54.3%) y β -cariofileno (28.9%), respectivamente. Se observó que el aceite esencial de *B. lanceolaria* era rico en hidrocarburos monoterpénicos (90.4%), mientras que los aceites esenciales de *H. indicum* y *T. rhomboidea* eran ricos en derivados de fenilo (72.8%) e hidrocarburos sesquiterpénicos (69.5%), respectivamente. Los compuestos comunes, a saber, terpin-4-ol y eugenol, se identificaron en los aceites esenciales de *B. lanceolaria*, *H. indicum* y *T. rhomboidea*.

Palabras clave: *Blumea lanceolaria* (Roxb.) Druce.; *Heliotropium indicum* L.; *Triumfetta rhomboidea* Jacq.; Terpin-4-ol; eugenol; GC / MS.

Introduction

“Herbal medicines are in widespread use and although many believe that herbal medicines are safe, and often used in combination are drawn from plant sources with their own variability in species, growing conditions, and biologically active constituents” [1]. “The essential oils are natural products that plants produce for their own needs other than nutrition (i.e. protection or attraction). In general, they are complex mixtures of organic compounds that give characteristic odor and flavor to the plants. They are mainly made up of monoterpenes and sesquiterpenes whose main metabolic pathway is through mevalonate leading to sesquiterpenes and from methyl-erythritol leading to monoterpenes” [2]. The exploration of traditional knowledge for the cure to common diseases is attractive since antiquity. The medicinal plants are responsible for most of the medicine and food used in modern society [3]. In India, about 70 percent of the rural population depends on the traditional Ayurvedic system of medicine. Most healers/practitioners of the traditional systems of medicine prepare formulations by their own recipes and dispense to the patients. The Western Ghats region is one of the 34 global biodiversity hotspots [4] which covers the Northwest Karnataka region.

Blumea lanceolaria (Roxb.) Druce. is belongs to the family Asteraceae. The leaves of *B. lanceolaria* are aromatic and used as a flavoring agent for the food. In traditional medicine, the leaves are used for the treatment of bronchitis, aphthae and asthma [5]. *B. lanceolaria* locally known as ‘Buarze’ in Mizoram, India and in folklore medicine this plant is used as an anti-cancer agent and leaves juice is useful for wound healing, chronic ulcers and infusion of leaves to control dysentery [6]. In the previous report, essential oil of *B. lanceolaria* has been reported a high amount of *p*-cymene [7] and methyl thymol [8] from India and Vietnam, respectively.

Heliotropium indicum L. of the family Boraginaceae is a high valued medicinal plant known in Ayurveda as Hastishundi Shrihastini or Vrishchikaali and distributed throughout India [9]. This is one of the most widely used herbs in the Ayurvedic medicine of India and some other countries for the treatment of liver diseases [10]. In traditional medicine, this plant is used as a local application for ulcers, wounds, sores, gum boils and skin affections [9]. The tribal community of Phulbani district of Odisha, India use the leaf paste of *H. indicum* over fresh cuts and wounds and claim for its promising activity [11] and scientifically reported wound healing activity on animals [11,12]. The decoction of leaves is used in urticaria and fevers and root is used for the treatment of coughs. Small doses of flower decoction are used for emmenagogue, while abortifacient in large doses [9]. The pharmacological activities like gastroprotective, antimicrobial, [13], anti-inflammatory [14], activities have been reported from various solvent extracts of *H. indicum*. The alkaloids *viz.*, indicine [10], indicinine [15], helindicine, lycopsamine [16], pestalamide B, glycynamide, N-(1-oxooctadecyl) glycy-L-alanylglycyl-L-histidyl [17] and indicine-N-oxide [18] have been isolated from *H. indicum*. The major volatile compounds phytol, 1-dodecanol, and β -linalool have been reported from Thailand and the oil showed significant anti-tuberculosis activity against *Mycobacterium tuberculosis* H37Ra in the Alamar blue assay system with a MIC of 20.8 μ g/mL [19]. Another report from Nigeria reported phenylacetaldehyde, (*E*)-2-nonenal and (*E,Z*)-2-nonadienal and hexahydrofarnesylacetone [20] were the major constituents.

Triumfetta rhomboidea Jacq. of the family Tiliaceae is commonly known in English as ‘Burbush’ or ‘Burweed’; in Ayurveda as ‘Jhinjhireetaa’ or ‘Gippit’ and in Karnataka ‘Bankitutturi’ and distributed throughout tropical and subtropical India, up to 1200 m in the Himalayas [9]. The plant is known as a weed, but a number of medicinal properties have been described for its different parts. In eastern Africa, *T. rhomboidea* is used in folk medicine for its abortive properties on the gravid mammalian uterus [21] and treatment of cancer among the tribal population in Kolli Hills, South India [22]. In traditional medicine, the bark and leaves of this plant are used for the treatment of diarrhea and dysentery [9]. Some pharmacopeia in eastern Africa described the uses of powdered leaves as an infusion for the treatment of anemia. In Kenya, this species is crushed and employed in massage to relieve labor pain. The infusion solution from its pounded roots is utilized as an anti-venom by applying to the bite area. In Rwanda, *T. rhomboidea* is used to treat pneumonia,

otitis, and diarrhea [23]. Various extracts of *T. rhomboidea* have been reported to possess cytotoxic [23], antioxidant [24] and antimicrobial [21] activities. A new compound triumfettamide B, along with lupeol, β -sitosterol, stigmasterol, oleanolic acid, maslinic acid, β -sitosterol glucopyranoside, and *trans*-tiliroside have been isolated and identified from *T. rhomboidea* [23]. The aerial parts of *T. rhomboidea* have been reported *trans*- β -caryophyllene, kessane and caryophyllene oxide as the major constituents [21].

This communication presents the essential oil composition of the different species of the traditional herbs viz., *B. lanceolaria*, *H. indicum* and *T. rhomboidea* (Fig. 1) growing Western Ghats region of India with the aim of possible utilization of the oil and demarcation of its terpenoid profile.

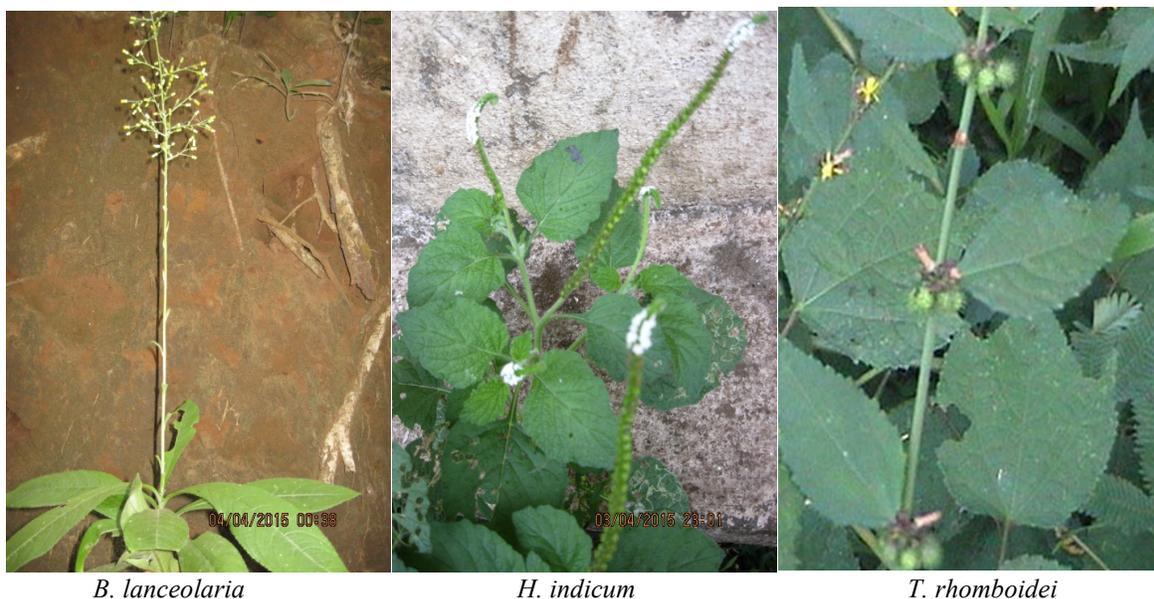


Fig. 1. Plants species of the traditional herbs growing in Western Ghats of North West Karnataka.

Experimental

Plant material

The leaves of *B. lanceolaria* and *H. indicum* were collected from Yana (N 14.51117; E 74.53061) forests of the Western Ghats in Uttara Kannada district, Karnataka, India, at a height of ~610 m. The aerial parts with raw seeds of *T. rhomboidea* were collected from Tilari of district Kholapur (N 15.79952; E 74.17441), Maharashtra, India, at a height of ~760 m. The plants were identified at ICMR-National Institute of Traditional Medicine (NITM), Belagavi, where the herbarium specimens have been deposited (*B. lanceolaria*: RMRC-1313; *H. indicum*: RMRC-1317; *T. rhomboidea*: RMRC-1333).

Isolation of essential oil

The dried leaves of *B. lanceolaria* (450 g), *H. indicum* (700 g) and fresh raw seeds of *T. rhomboidea* (250 g) were hydro-distilled separately for 3 h using a Clevenger type apparatus. The obtained oil was dried over anhydrous sodium sulfate and stored at -4°C until further analysis. The yield of the oil was 0.02% w/w for *B. lanceolaria*; 0.01% w/w for *H. indicum* and 0.15% w/w for *T. rhomboidea*.

Chromatographic analysis

The chemical composition of the essential oils of *B. lanceolaria*, *H. indicum*, and *T. rhomboidea* (1% solution of essential oil in equal ratio of *n*-hexane: dichloromethane) were analyzed by using a Gas Chromatograph (GC) (Varian 450 fitted with a fused silica capillary column BP-1 (100% dimethyl polysiloxane; SGE Analytical Science), 30 m × 0.25 mm i.d., 0.25 μm film thickness) under the experimental conditions reported earlier [25-27]. The oven temperature was programmed from 60-220°C at 3°C/min, using nitrogen as a carrier gas. The injector and the detector (FID) temperature were set at 230°C and 240°C, respectively. Gas Chromatography-Mass Spectrometer (GC-MS) analysis was employed as a Thermo Scientific Trace Ultra GC interfaced with a Thermo Scientific ITQ 1100 mass spectrometer fitted with a BP-1 (100% dimethylpolysiloxane; SGE Analytical Science) fused silica capillary column (30 m × 0.25 mm; 0.25 μm film thickness). The oven temperature range was programmed from 60-220°C at 3°C/min and helium was used as carrier gas at 1.2 mL/min for analysis. The injector temperature was set at 230°C, and the injection volume was 0.1 μL in *n*-hexane, with a split ratio of 1:50. MS were taken at 70 eV with a mass range of *m/z* 40-450 and other parameters used were those reported earlier [28-30]. Identification of constituents were done on the basis of Retention Index (RI, determined [Linear Temperature programmed analysis was used with the formula $RI = 100 \times [(t_{Ri} - v) - (t_{RZ} - v) / (t_{R(Z+1)} - v) - (t_{RZ} - v) + Z]$ where t_{Ri} = retention time of sample peak; v = column void time; t_{RZ} = retention time of *n*-alkane peak eluting immediately before sample peak; $t_{R(Z+1)}$ = retention time of *n*-alkane peak eluting immediately after sample peak; Z = carbon number of *n*-alkane peak eluting immediately before sample peak [31] with reference to homologous series of *n*-alkanes C₈-C₂₅ under identical experimental conditions on BP-1 column), MS library search NIST 08 MS Library (Version 2.0 f; Thermo Fisher Scientific Austria) and WILEY MS 9th Edition (Thermo Fisher Scientific Austria), and by comparing with the MS literature data and co-injection of commercial samples from Sigma-Aldrich, India (≥98% purity). The relative amounts of individual components were calculated based on the GC peak area (FID response) without using a correction factor.

Results and discussion

Chemical components of essential oils from *B. lanceolaria*, *H. indicum*, and *T. rhomboidea* have been presented in Table 1 in elution order from the BP-1 column, along with the percentage composition of each component and its retention index. Thirty-six, twenty-seven, and fifty constituents from the essential oils of *B. lanceolaria*, *H. indicum*, and *T. rhomboidea*, respectively were identified according to their mass spectra and their relative retention indices determined in a nonpolar stationary phase capillary column. The essential oils of *B. lanceolaria*, *H. indicum*, and *T. rhomboidea* comprising 97.3%, 98.2%, and 97.6%, respectively of the total oil constituents. The major compound of the essential oil of *B. lanceolaria* was identified as β-pinene (82.3%) along with other minor constituents terpin-4-ol (4.1%), γ-terpinene (2.5%) and sabinene (2.2%). The oil was found to be rich in monoterpene hydrocarbons (90.4%), followed by oxygenated monoterpenes (6.5%) and phenyl derivatives (0.2%). The major constituent of the essential oil of *H. indicum* was methyl salicylate (54.3%). The other minor constituents were sabinene (7.9%), eugenol (6.3%) and *cis*-calamene (4.7%). The oil was found to be rich in phenylpropanoid type constituents (72.8%), followed by monoterpene hydrocarbons (17.6%), sesquiterpene hydrocarbon (6.6%), oxygenated monoterpene (0.9%) and oxygenated sesquiterpene (0.3%). The major compound of the essential oil of *T. rhomboidea* was identified as β-caryophyllene (28.9%). The other minor constituents were α-humulene (8.5%), germacrene D (6.9%), *cis*-calamene-10-ol (6.5%), α-cadinol (5.4%) and δ-cadinene (4.8%). The oil was found to be rich in sesquiterpene hydrocarbons (69.5%), followed by oxygenated sesquiterpenes (21.5%), oxygenated long-chain hydrocarbons (3.7%), monoterpene hydrocarbon (1.6%), and phenyl derivatives (1.3%) type constituents. The compounds terpin-4-ol (0.1-4.1%) and eugenol (t-6.3%) have been commonly identified in the essential oils of *B. lanceolaria*, *H. indicum*, and *T. rhomboidea*.

Table 1. Chemical composition of the essential oil of *B. lanceolaria* *H. indicum* and *T. rhomboidea*.

Compound	RT	RI	% Content			Identification *
			<i>B. lanceolaria</i>	<i>H. indicum</i>	<i>T. rhomboidea</i>	
Tricyclene	5.33	931	0.1	-	-	RI, MS
α -Thujene	5.78	944	0.1	1.1	-	RI, MS
α -Pinene	6.04	951	1.1	2.3	-	RI, MS, CI
Camphene	6.38	961	0.1	-	-	RI, MS
Sabinene	6.9	976	2.2	7.9	-	RI, MS
β -Pinene	7.05	980	82.3	-	-	RI, MS
<i>cis-m</i> -Mentha-2,8-diene	7.26	986	0.1	-	-	RI, MS
Myrcene	7.49	993	-	3.8	-	RI, MS, CI
<i>m</i> -Mentha-1(7),8-diene	7.55	995	t	-	-	RI, MS
α -Phellandrene	7.95	1006	0.5	-	-	RI, MS, CI
α -Terpinene	8.33	1017	0.6	0.5	-	RI, MS
<i>p</i> -Cymene	8.42	1019	t	t	-	RI, MS
Limonene	8.73	1028	0.1	-	-	RI, MS, CI
1,8-Cineole	8.74	1029	-	0.7	-	RI, MS, CI
(<i>Z</i>)- β -Ocimene	8.98	1035	0.1	-	-	RI, MS
(<i>E</i>)- β -Ocimene	9.37	1047	0.1	-	-	RI, MS
γ -Terpinene	9.73	1057	2.5	1.1	-	RI, MS, CI
<i>cis</i> -Sabinene hydrate	9.9	1062	0.5	0.2	-	RI, MS
<i>m</i> -Cymenene	10.61	1082	t	-	-	RI, MS
Terpinolene	10.83	1088	0.5	0.9	-	RI, MS, CI
Linalool	10.99	1093	0.1	-	-	RI, MS, CI
<i>trans</i> -Sabinene hydrate	11.05	1095	0.6	-	-	RI, MS
<i>cis</i> -Decahydro naphthalene	11.1	1096	-	0.3	-	RI, MS
<i>cis</i> -Thujone	11.14	1097	-	-	t	RI, MS
1,3,8- <i>p</i> -Menthatrien	11.16	1098	t	-	-	RI, MS
dehydro Sabina ketone	11.55	1109	-	-	0.1	RI, MS
<i>cis-p</i> -Menth-2-en-1-ol	11.91	1119	0.3	-	-	RI, MS
<i>trans-p</i> -Menth-2en-1-ol	12.54	1137	0.1	-	-	RI, MS
<i>trans</i> -Verbenol	12.75	1143	0.1	-	-	RI, MS
Menthofuran	13.53	1165	-	2.1	-	RI, MS
Terpin-4-ol	14.12	1182	4.1	1.9	0.1	RI, MS, CI
α -Terpineol	14.33	1188	-	-	0.2	RI, MS, CI
Methyl salicylate	14.56	1195	-	54.3	-	RI, MS
Methyl chavicol	14.66	1198	-	-	0.3	RI, MS, CI
<i>cis</i> -Piperitol	14.84	1203	0.2	-	-	RI, MS, CI
<i>trans</i> -dihydrocarvone	15.18	1213	-	-	1.2	RI, MS
<i>trans</i> -Piperitol	15.28	1215	0.1	-	-	RI, MS, CI
(<i>Z</i>)-Ocimenone	16.05	1237	-	2.4	-	RI, MS
Thymol methyl ether	16.11	1239	t	-	-	RI, MS
Pulegone	16.21	1242	t	-	-	RI, MS, CI
(<i>E</i>)-Ocimenone	16.35	1246	-	2.6	-	RI, MS
Cumin aldehyde	16.54	1251	0.2	-	-	RI, MS
Carvone	16.85	1260	0.1	-	-	RI, MS, CI
(<i>E</i>)-Anethole	18.21	1299	t	t	-	RI, MS, CI

Isobornyl acetate	18.36	1303	-	-	t	RI, MS
Thymol	18.58	1310	0.3	t	-	RI, MS, CI
<i>p</i> -Cymen-7-ol	18.72	1314	t	-	-	RI, MS
Carvacrol	19.07	1324	0.2	t	-	RI, MS, CI
Eugenol	21.07	1381	t	6.3	1.0	RI, MS, CI
Cyclosativene	22.54	1423	-	-	0.8	RI, MS
Isoledene	22.67	1427	-	-	t	RI, MS
α -copaene	22.87	1432	-	-	0.3	RI, MS
β -Bourbonene	23.08	1438	-	-	0.1	RI, MS
β -Elemene	23.48	1450	-	-	0.3	RI, MS
Methyl eugenol	23.8	1459	-	1.2	-	RI, MS, CI
Longifolene	23.99	1464	-	-	0.5	RI, MS
α -Gurjunene	24.24	1471	-	-	0.8	RI, MS
β -Caryophyllene	24.68	1484	-	0.8	28.9	RI, MS, CI
β -Gurjunene	24.88	1490	-	-	1.7	RI, MS
γ -Elemene	25.06	1495	-	-	0.7	RI, MS
6,9-Guaiadene	25.42	1505	-	-	3.6	RI, MS
<i>cis</i> -Muurolo-3,5-diene	25.63	1511	-	-	0.3	RI, MS
α -Humulene	25.77	1515	-	t	8.5	RI, MS
Sesquisabinene	26.06	1523	-	-	2.8	RI, MS
4,5-di- <i>epi</i> -Aristolochene	26.5	1536	-	-	0.9	RI, MS
γ -Gurjunene	26.61	1539	-	-	0.3	RI, MS
(<i>E</i>)- β -Ionone	26.73	1543	-	1.7	-	RI, MS
β -Selinene	27.04	1551	-	-	2.6	RI, MS
<i>trans</i> -Muurolo-4(14), 5-diene	27.19	1556	-	-	1.7	RI, MS
<i>epi</i> -Cubebol	27.35	1560	-	-	1.2	RI, MS
α -Selinene	27.45	1563	-	-	0.3	RI, MS
α -Muurolo-4	27.65	1569	-	-	0.5	RI, MS
Germacrene A	27.77	1572	-	-	0.3	RI, MS
γ -Cadinene	28.09	1581	-	-	1.1	RI, MS
<i>cis</i> -Calamenene	28.21	1585	-	4.7	-	RI, MS
δ -Cadinene	28.25	1586	-	1.1	4.8	RI, MS
<i>trans</i> -Calamenene	28.46	1592	-	-	t	RI, MS
<i>trans</i> -Cadin-1,4-diene	28.8	1602	-	-	0.2	RI, MS
α -Cadinene	28.88	1604	-	-	6.9	RI, MS
α -Calacorene	28.93	1605	-	-	0.5	RI, MS
Germacrene B	29.28	1615	-	-	1.4	RI, MS
<i>cis</i> -Muurolo-5-en-4- α -ol	29.44	1620	-	-	-	
Germacrene D-4-ol	30.28	1644	-	-	1.0	RI, MS
Caryophyllene oxide	30.37	1647	-	0.3	2.2	RI, MS, CI
<i>epi</i> -Cedrol	31.54	1680	-	-	0.7	RI, MS
1,10-di- <i>epi</i> -Cubenol	31.72	1685	-	-	0.1	RI, MS
1- <i>epi</i> -Cubenol	32.18	1698	-	-	0.2	RI, MS
<i>epi</i> - α -Cadinol	32.65	1712	-	-	1.6	RI, MS
Cubenol	32.75	1715	-	-	1.2	RI, MS
α -Cadinol	33.09	1724	-	-	5.4	RI, MS
<i>cis</i> -Calamene-10-ol	33.26	1729	-	-	6.5	RI, MS
Zizanal	34.39	1761	-	-	t	RI, MS
14-hydroxy- α -Humulene	35.32	1788	-	-	t	RI, MS

Hexadecanoic acid	43.99	2036	-	-	2.9	RI, MS, CI
Methyl linoleate	48.86	2175	-	-	0.8	RI, MS, CI
Monoterpene hydrocarbons			90.4	17.6		1.6
Oxygenated monoterpenes			6.7	0.9		69.5
Sesquiterpene hydrocarbons			-	6.6		21.5
Oxygenated sesquiterpene			-	0.3		1.3
Phenyl derivatives			0.2	72.8		1.3
Oxygenated long chain hydrocarbons			-	-		3.7
Total identified			97.3	98.2		97.6

RT=Retention Time, *RI=Retention index relative to C₈-C₂₅ n-alkanes on BP-1 column, MS=NIST and Wiley library and the literature, t=trace (<0.1%), CI=Co-injection of commercial samples.

The essential oil constituents of *B. lanceolaria* growing in the Western Ghats region of India has somehow resembled the plant grown in Vietnam and other regions of India. The chemical constituents of the plant *B. lanceolaria* from Vietnam viz., α -thujene (0.04%), α -pinene (0.01%), camphene (0.01%), β -pinene (0.01%), α -terpinene (0.01%), *p*-cymene (3.28%), limonene (0.12%), methyl thymol (94.96%), and β -caryophyllene (0.04%) have been common to this report in quantitative divergence and methyl carvacrol (0.02%) and 1-hexadecanol (0.79%) which were reported from Vietnam [8] are not detected in this report. The rest of the compounds identified in this report are not identified in Vietnam grown *B. lanceolaria* (Table 1). Similarly, the only compound *p*-cymene (99%) has been reported from India in the very high amount [7] and this report this compound has been identified in trace (<0.1%).

A comparison of essential oil constituents of *H. indicum* of this study with the compounds of plant growing in Thailand [19] was found to be completely different. Apart from this, the report from Nigeria grown *H. indicum* plant only three compounds viz., methyl salicylate (1.2%), (*E*)- β -ionone (1.9%) and eugenol (2.4%) [20] were found to be common compared to the present study.

The essential oil constituents reported [21] from the aerial parts of *T. rhomboidea* in terms of the major compound was similar to that found in this study with quantitative differences, while qualitatively relative amounts of the components which are somehow dissimilar from France growing plant. Correlation of the compounds from France growing plant with this study revealed that a high amount of β -caryophyllene and low amount of caryophyllene oxide were found in raw seeds oil of *T. rhomboidea*. It is interesting to note that the compound kessane which was the second major compound from France growing *T. rhomboidea* could not be detected even trace amount in the raw seed oil in this study. Overall, out of fifty compounds identified in the raw seeds of *T. rhomboidea*, twenty-one compounds were found to be similar with varying quantities as compared to the aerial parts of France growing *T. rhomboidea* plant [21]. Interestingly, the cumulative quantity of twenty-one similar constituents of raw seeds and aerial parts contains more or less similar quantities (65.4% from *T. rhomboidea* raw seeds oil and 65.7% from aerial parts *T. rhomboidea* oil.

The compound β -pinene possesses various biological activities viz., anti-infectious bronchitis virus [32], antinociceptive [33], antimicrobial [34] as well as antiviral against herpes simplex virus [35] and β -pinene present in high amount in the leaves of *B. lanceolaria* may support the traditional uses of this herb for the treatment of bronchitis, aphthae, asthma [5], wound healing, chronic ulcers, and to control dysentery [6].

The compound methyl salicylate in a small amount has been used as a topical counterirritant for the treatment of mild aches and pains of arthritis, as well as for acute muscle strain associated with the “weekend warrior” syndrome [36]. Hence, the plant *H. indicum* used as local application support traditional medicine.

The β -caryophyllene is the main compound of the essential oil of *T. rhomboidea* and β -caryophyllene has anti-inflammatory [37], anticancer [38], antinociceptive [39], cytoprotective [40], analgesic [38,41] and showed marked protective [42] effects. The β -caryophyllene is an excellent therapeutic agent and the use of *T. rhomboidea* in traditional medicine may attribute to the treatment of various ailments like the treatment of cancer, diarrhea, gonorrhoea to stop the burning caused by urine, treatment of anemia, relieve labor pain, etc.

The quantitative and qualitative divergence may be due to the geographical, climatic, and soil conditions, which in turn may affect the composition and other secondary metabolites of the plants [43,44]. Essential oil formation in the plants is highly dependent on climatic conditions, especially day length,

irradiance, temperature, and water supply. Tropical species follow in their vegetation cycle the dry and rainy seasons, while species of the temperate zones react more on day length, the more distant from the equator their natural distribution area is located [45]. The ecological experiments regard to the essential oil content was found a correlation between flower formation, flowering period, and essential oil synthesis [45]. It was observed that the shorter the flowering phase, the less was the time available for oil formation, and thus the lower oil content. The composition of the essential oil, on the other hand, showed no qualitative change due to ecological or climatic factors confirming that chemotypes keep their typical pattern [45].

Conclusion

The present investigation concludes that the essential of *H. indicum* collected from south India was found to be methyl salicylate chemotype. Hence, *H. indicum* could be a good source of methyl salicylate, which is the main constituent of the oil. In conclusion, the essential oil constituents vary in plant parts as well as in different localities growing plants. The plant *T. rhomboidea* is a high valued medicinal plant and as per the geoclimatic distribution of the flora, botanicals are consumed by the people as their own localities to cure the diseases. Secondary metabolites of the plants are changes in quantity or produced different chemotypes which may alter the biological activities. This study revealed that qualitative and quantitative difference in case of *B. lanceolaria* oil and compared from the previous reports, could exhibit altered bioactivities. Further studies are required to investigate the biological activities of the volatile constituents of these plants.

Acknowledgments

The author is grateful to the Indian Council of Medical Research (ICMR), New Delhi, India for providing the necessary facilities, and is thankful to Mr. Mahesh B. Wagarwadi, Multi Tasking Staff (MTS), Department of Phytochemistry, ICMR-NITM, Belagavi for processing and extraction of the oils.

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