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Chemical composition of the essential oil of *Euphorbia pilosa* from Munsiari, Pithoragarh, India

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Abstract
Introduction: *Euphorbia pilosa* is much branch, milky latex bearing tall herb commonly used to treat bronchitis, Purgative, emetic, fistulous sores and asthma-related problems. The plants were collected in the month of September 2015 from Kalamuni (Munsiari) near Pithoragarh, location of Kumaon Himalayas.
Methods: The plant *Euphorbia pilosa* including leaves, stem, and flowers were extracted by hydro distillation method for 6 hours using Clevenger apparatus. The hydro-distilled essential oil of *Euphorbia pilosa* has been examined by means of gas chromatography-mass spectrometry (GC-MS).
Results: Phytochemical investigation of whole plants of *Euphorbia pilosa* led to the isolation and identification of essential oil. Separation was achieved by Elite-5MS fused capillary column. The mass spectra were compared with the spectra of known components stored in the NIST and WILEY databases for compound identification. Forty-six chemical constituents were identified. The major constituents were Phytol (5.75%), n-Pentadecanal (5.12%), n-Pentadecane (4.02 %), δ-Terpineol (3.31) and Tricos-(9Z)-ene (3.24%).
Conclusion: The results data obtained in the present study suggest that essential oil possesses strong medicinal activities can be utilized for treatment of diseases.

Keywords: phytol, δ-terpineol, *Euphorbia pilosa*, essential oil, GC-MS

1. Introduction

The genus Euphorbia is the largest in the spurge family, comprising more than 2000 species (Jassbi, 2006) [3]. Some species of Euphorbia have been used as medicinal plants for the treatment of skin diseases, gonorrhea, migraine, and intestinal parasites, and as wart cures (Singla 1990) [6]. Roots have laxative and emetic properties. Euphorbia pilosa is a much branched, milky latex bearing tall herb of 40-80 cm. Leaves are alternate, elliptic-oblong to narrow elliptic. Inflorescence with flowers in heads having cup shaped bracts which encircles the male flowers, each with one stamen that surround a single female flower. Female flowers are three chambered, stalked ovary. Phytochemical investigation of whole plants of Euphorbia pilosa led to the isolation and identification of two new daphnane-diterpenoid glucosides, euphopilosides A and B (1 and 2, resp.), and a new ent-abietane, euphopilolide (3), together with eight known compounds (Zhang, 2014) [8]. The plant's sap is toxic to rapidly replicating human tissue, and has long been used as a traditional remedy for common skin lesions, including cancer (Siller *et al*., 2009) [5].

Medicinal plants have always an important place in the therapeutic system. The use of natural products in the treatment of various diseases has played an important role in medical therapy for many years and plants of the genus Euphorbia are known to possess considerable medicinal and economic importance components (Tian *et al*., 2010 and Chaabi *et al*., 2007) [7, 2].

This is the first work of chemical composition of essential oil of *Euphorbia pilosa* higher altitudes of Kumaon Himalayas.

2. Materials and Methods

2.1 Plant Material

The leaves of *Euphorbia pilosa* were collected in the month of September 2015 from September 2015 from Kalamuni (Munsiari) near Pithoragarh, India in the Kumaon Himalayas. The plant was authenticated by Botanical Survey of India (BSI) and identification no. was 114845.

2.2 Isolation of essential oil

The plant *Euphorbia pilosa* including leaves, stem, and flower was extracted by hydro-distillation method for 6 hours using Clevenger apparatus. The oil was dried over anhydrous sodium sulphate and stored at room temprature in a sealed vial until analysis was performed. The percentage oil yield was calculated based on the dry weight of the plant. The oil yield was (0.08%).

2.3 GC and GC/MS analyses and identification

Essential oil analyses were performed by GC-MS and GC-FID on a Shimadzu QP-2010 instrument, equipped with FID, in the same conditions. The percentage composition of the oil samplewas computed from the GC peak areas without using correction for response factors. The oilwas analyzed using a Shimadzu GC/MS Model QP 2010 Plus, equipped with a Rtx-5MS (30 m ×0.25 mm; 0.25 mm film thickness) fused silica

capillary column. Helium (99.999 %) was used as a carrier gas adjusted to 1.21 ml/min at 69.0 K Pa, splitless injection of 1 mL, of a hexane solution injector and interface temperature were 270 ^{0}C, oven temperature programmed was 50–280 ^{0}C at 3 ^{0}C/min. Mass spectra were recorded at70 eV. Ion source temperature was 230 ^{0}C.

The identification of the chemical constituents was assigned on the basis of comparison of their retention indices and mass spectra with those given in the literature (Adams, 2001) [1]. Retention indices (RI) were determined with reference to a homologous series of normal alkanes, by using the following formula (Kovats, 1958) [4].

$$KI = 100\,[n + (N-n) \times \frac{\log t^1_R\,(\text{unknown}) - \log t^1_R\,(C_n)}{\log t^1_R\,(C_N) - \log t^1_R\,(C_n)}$$

t^1_R – the net retention time ($t_R - t_0$)
t_0 – the retention time of solvent (dead time)
t_R – the retention time of the compound.
C_N – number of carbons in longer chain of alkane
C_n– number of carbons in shorter chain of alkane
n - is the number of carbon atoms in the smaller alkane
N - is the number of carbon atoms in the larger alkane

3. Results and Discussion

The GC and GC-MS analyses of essential oil of *Euphorbia pilosa* resulted in the identification of eight one compounds (table-1). The oil yield was found to be 0.08 % by weight. Both, the major as well as minor constituents were identified by their retention indices and comparison of their mass spectra. Total seventy-seven compounds were identified constituting 81.59 % of the total oil. Phytochemical investigation of whole plants of Euphorbia pilosa led to the isolation and identification of essential oil. Separation was achieved by Elite-5MS fused capillary column. The mass spectra were compared with the spectra of known components stored in the NIST and WILEY databases for compound identification. Forty-six chemical constituents were identified. The major constituents were Phytol (5.75%), n-Pentadecanal (5.12%), n-Pentadecane (4.02 %), δ-Terpineol (3.31) and Tricos-(9Z)-ene (3.24%). The main minor compounds were 2,3,4-Trimethylpentane (0.10%), E-2-Tetradecen-1-ol (0.13%), 2,3,7-Trimethyloctane (0.16%), γ-Elemene (0.16 %), epi-Cedrol (0.16%), Lilac aldehyde B (0.17%) and Methyl(2E)-6-isopropyl-4-(1-methylethylidene)-2,6-heptadienoate (0.17%).

Table 1: Essential oil composition of *Euphorbia pilosa*

S.N.	Compound Name	Area%	Mol. Formula	Mol. Weight	Retention Indices	Mode of Identific-ation
1	2,3,4-Trimethylpentane	0.10	C_8H_{18}	114	624	
2	2,4-Dimethylhexane	0.96	C_8H_{18}	114	688	
3	Diisobutylene	0.42	C8H16	112	717	
4	n-Octane	1.19	C_8H_{18}	114	800	
5	2-Ethyl-5-methyltetrahydrofuran	1.04	$C_7H_{14}O$	114	810	
6	Ethylcyclohexane	0.26	C_8H_{16}	112	880	
7	isopropyl-Cyclohexane	0.37	C_9H_{18}	126	915	
8	n-Nonane	0.62	C_9H_{20}	128	916	
9	2,3,7-Trimethyloctane	0.16	$C_{11}H_{24}$	156	922	
10	n-Decane	1.16	$C_{10}H_{22}$	142	1015	
11	3,4-Dimethyl decane	0.52	$C_{12}H_{26}$	170	1086	
12	β-Linalool	1.60	$C_{10}H_{18}O$	154	1086	
13	n-Undecane	1.68	$C_{11}H_{24}$	156	1115	
14	1-Methylbutyl-cyclohexane	0.31	$C_{11}H_{22}$	154	1116	
15	δ-Terpineol	3.31	$C_{10}H_{18}O$	154	1170	
16	Terpinen-4-ol	0.38	$C_{10}H_{18}O$	154	1180	
17	5,6-Dimethylundecane	0.40	$C_{13}H_{28}$	184	1185	
18	2-Methyl-5-propylnonane	1.59	$C_{13}H_{28}$	184	1185	
19	6-Ethyl-2-methyldecane	0.32	$C_{13}H_{28}$	184	1186	
20	Hotrienol	3.22	$C_{10}H_{18}O_2$	170	1197	
21	Lilac aldehyde B	0.17	$C_{10}H_{16}O2$	168	1198	
22	n-Dodecane	0.24	$C_{12}H_{26}$	170	1200	
23	Pseudocyclocitral	0.18	$C_{10}H_{16}O$	152	1204	
24	2-(2-methylpropylidene) Cyclohexanone	0.20	$C_{10}H_{16}O$	150	1209	
25	Fragranol	0.41	$C_{10}H_{18}O$	154	1212	
26	5-Butylnonane	0.26	$C_{13}H_{28}$	184	1249	
27	Linalyl acetate	3.19	$C_{12}H_{20}O_2$	196	1250	
28	Geraniol	0.49	$C_{10}H_{18}O$	154	1255	
29	Dec-(2E)-enal	0.42	$C_{10}H_{18}O$	154	1265	
30	4-ethyl-Guaiacol	2.55	$C_9H_{12}O_2$	152	1275	
31	n-Tridecane	1.47	$C_{13}H_{28}$	184	1300	
32	Cyclosativene	0.49	$C_{15}H_{24}$	204	1364	
34	Undec-(8Z)-enal	0.47	$C_{11}H_{20}O$	168	1365	
35	Copaene <alpha->	0.90	$C_{15}H_{24}$	204	1375	

36	β-elemene	0.38	$C_{15}H_{24}$	204	1390	
37	n-Tetradecane	1.47	$C_{14}H_{30}$	198	1400	
38	α-Cedrene	0.37	$C_{15}H_{24}$	204	1414	
39	(E)-Caryophyllene	1.66	$C_{15}H_{24}$	204	1424	
40	γ-Elemene	0.16	$C_{15}H_{24}$	204	1432	
41	γ-Muurolene	0.54	$C_{15}H_{24}$	204	1435	
42	Methyl(2E)-6-isopropyl-4-(1-methylethylidene)-2,6-heptadienoate	0.17	$C_{14}H_{22}O_2$	222	1453	
43	α-Humulene	0.42	$C_{15}H_{24}$	204	1454	
44	β-Ionone	0.53	$C_{13}H_{20}O$	192	1468	
45	Selina-4,11-diene	0.17	$C_{15}H_{24}$	204	1476	
46	β-Chamigrene	0.51	$C_{15}H_{24}$	204	1480	
47	Germacrene D	1.33	$C_{15}H_{24}$	204	1480	
48	n-Pentadecane	4.02	$C_{15}H_{32}$	212	1500	
49	(E,E)- α-Farnesene	0.44	$C_{15}H_{24}$	204	1504	
50	γ-Cadinene	0.26	$C_{15}H_{24}$	204	1510	
51	δ-Cadinene	1.04	$C_{15}H_{24}$	204	1520	
52	(6E)-Nerolidol	0.30	$C_{15}H_{26}O$	222	1564	
53	Caryophyllene oxide	0.55	$C_{15}H_{24}O$	220	1584	
54	n-Hexadecane	0.94	$C_{16}H_{34}$	226	1600	
55	Cedrol	0.66	$C_{15}H_{26}O$	222	1603	
56	epi-Cedrols	0.16	$C_{15}H_{26}O$	222	1621	
57	Epicubenol	0.79	$C_{15}H_{26}O$	222	1631	
58	δ-Cadinol	0.85	$C_{15}H_{26}O$	222	1641	
59	Himachalol	0.19	$C_{15}H_{26}O$	222	1650	
60	neo-Intermedeol	0.59	$C_{15}H_{26}O$	222	1661	
61	E-2-Tetradecen-1-ol	0.13	$C_{14}H_{28}O$	212	1664	
62	Hedycaryol	0.26	$C_{15}H_{26}O$	222	1694	
63	n-Heptadecane	0.99	$C_{17}H_{36}$	240	1700	
64	1-Heptadecene	1.75	$C_{17}H_{34}$	238	1701	
65	n-Pentadecanal	5.12	$C_{15}H_{30}O$	226	1702	
66	Longifolol	1.82	$C_{15}H_{26}O$	222	1715	
67	Hexahydrofarnesyl acetone	1.63	$C_{18}H_{36}O$	268	1754	
68	n-Pentadecanol	0.47	$C_{15}H_{32}O$	228	1755	
69	(11E)-12-Cyclopropyl-11-dodecen-1-ol	0.63	$C_{15}H_{28}O$	224	1765	
70	cis,cis,cis-7,10,13-Hexadecatrienal	1.78	$C_{16}H_{26}O$	234	1824	
71	Nonadec-1-ene	1.12	$C_{19}H_{38}$	266	1900	
72	Isophytol	0.30	$C_{20}H_{40}O$	296	1948	
73	Pimaradiene	0.22	$C_{20}H_{32}$	272	1953	
74	3.alpha.-dihydronandrolone	0.49	$C_{18}H_{28}O_2$	276	2080	
75	Phytol	5.75	$C_{20}H_{40}O$	296	2114	
76	Nonadecyl alcohol	1.21	$C_{19}H_{40}O$	284	2150	
77	n-Docosane	1.95	$C_{22}H_{46}$	310	2200	
78	3-Ethyl-3-hydroxyandrostan-17-one	0.46	$C_{21}H_{34}O_2$	318	2251	
79	Tricos-(9Z)-ene	3.24	$C_{23}H_{46}$	322	2274	
80	3-alpha-hydroxy-Manool	0.45	$C_{20}H_{34}O_2$	306	2296	
81	n-Pentacosane	1.13	$C_{25}H_{52}$	352	2500	
82	n-Octyl phthalate	0.23	$C_{24}H_{38}O_4$	390	2832	
83	n-Tetratriacontane	0.91	$C_{34}H_{70}$	478	3400	
84		81.59				

4. Conclusions

The essential oil from *Euphorbia pilosa* showed a qualitative and quantitative make-up of constituents. Clinically, this herb can be a good source of herbal medicine for the treatment of diseases indigenously. The study will also help to generate a database of species which can be exploited scientifically and judiciously in the future by local people and so that ecological balance is maintained. The results obtained in the present study suggest that the essential oil of *Euphorbia pilosa* possesses medicinally active compounds.

This is the first report of chemical composition of essential oil of *Euphorbia pilosa* higher altitudes of Kumaon Himalayas.

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Author Contributions

The first author, Chandan Ram, who pursues his Ph.D under the supervision of Prof Pushpa Joshi, carried out all the

experimental work. Kundan Prasad, the third author, designed all the experiments, analyzed the data, and prepared the manuscript. All authors read and approved the final manuscript.

Conflicts of Interest

The authors declare that there is no conflict of interest.

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