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CHEMICAL COMPOSTION OF ESSENTIAL OIL OF *EXCOECARIA ACERIFOLIA* COLLECTED FROM KUMAUN HIMALAYAS

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ABSTRACT

The hydro-distilled essential oil of *Excocaria acerifolia* has been examined by means of gas chromatography-mass spectrometry (GC-MS). The oil constituents were identified according to their mass spectra and their relative retention indices determined on a non-polar stationary phase capillary column. Fifty-seven constituents have been identified representing 93.37% of the total oil. The main compounds in major amounts were Isopropyl isothiocyanate (14.03%), (E)-Caryophyllen (11.65%), Benzyl isothiocyanate (9.32%), 1, 3, 8- para-menthatriene (6.61%), the compound in trace or in minor amount were n-hexanol (0.06%), 2-Ethyl-5-methyl tetrahydrofuran (0.06%), the oil was found rich in oxygenated mono and sesquiterpene hydrocarbons.

KEYWORDS: (E)-caryophyllene, Euphorbiaceae, terpenoid composition, Essential oil, GC-MS Mass spectroscopy.

INTRODUCTION

Euphorbiaceae is a large family of flowering plants with 300 genera and around 7,500 species. Most spurges are herbs, but some, especially in the tropics, are shrubs or trees. Some are succulent and resemble cacti (Ahmed *et al*., 2007-2009). The genus can be found all over the world. A milky latex is a characteristic of the subfamilies *Euphorbiaceae* and *Crotonoideae as well*. This latex is poisonous in the *Euphorbioideae*. In China, there are five recorded *Excoecaria* species, of which *Excoecaria agallocha* has been well studied and a number of novel diterpenoids with anti-tumor bioactivity (Tenji *et al*.,2000) and anti HIVphorbol ester (Anjaneyulu *et al*., 2003) found in it have been reported. However, no information concerning the chemical constituents of *E. acerifolia* has been published so far

The *E. acerifolia* plant is a dominant species of Maquis vegetation type distributed in the dry valleys in Southwest China (Wu *et al*., 2003 & Jin *et al*., 1998).

Among the plants often used in traditional medicine, *Excoecaria* species, which belong to the *Euphorbieacea* family, play a vital role. A number of plants of the spurge family are of considerable economic importance. Leafy spurge (*Euphorbia esula*) and Chinese tallow (*Triadica sebifera*) are invasive weeds in North America. In medicine, some species of *Euphorbiaceae* have proved effective against genital herpes (Charles *et al*., 2007). Essential oils are valuable natural products, which are used as raw materials in a various way including perfumes, cosmetics, aromatherapy, phytotherapy, spices and nutrition (Buchbauer, 2000).

Experimental

Plant material

The leaves of *Excocaria acerifolia* were collected in the month of August 2015 from Pithoragrah near APS School location of Kumaon Himalayas, mainly The plant was authenticated by Botanical Survey of India (BSI).

Essential oil extraction

The leaves of *Excocaria acerifolia*, were extracted by hydro distillation method for 8 hours using Clevenger apparatus ((Anonymous *et al*., 1996). with 1000 g of leaves of each sample. *Excocaria acerifolia*, the oil yield was not so good, The oil was dried with 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 leaf. The oil yield were(0.12%),.

GC-FID and GC-MS analysis

Essential oil analysis was performed by using GC-MS and GC-FID was performed on a Shimadzu QP-2010 instrument, equipped with FID, in the same conditions, except hydrogen was used as the carrier gas. The percentage composition of the oil samples were computed from the GC peak areas without using correction for response factors. The oils were 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 was used as carrier gas adjusted to 1.21 ml/min at 69.0 KPa; splitless injection of 1 mL, of a hexane solution; injector and interface temperature were 270oC; oven temperature programmed was 50–280oC at 3 C/min. EIMS: electron energy, 70 eV; ion source temperature was 230^{o}C.

Identification of constituents were done on the basis of Retention Index (RI, determined with reference to homologous series of n-alkanes C8-C28, under identical experimental condition), MS library search (NIST and WILEY), and by comparison with MS literature data (Adams *et al.*, 2001). The relative amounts of individual components were calculated based on GC peak area (FID response) without using correction factor. Retention indices (RI) were determined with reference to a homologous series of normal alkanes, by using the following formula (Kovats *et al*,, 1958).

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

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

RESULT AND DISCUSSION

The GC and GC-MS analysis of leaf oil of *Excoecaria acerifolia* resulted in the identification of fifty-seven constituents, the oil yield was found to be 0.5 % by weight. Both, the major as well as minor constituents were identified by their retention indices and comparison of their mass spectra. Total fifty-seven constituents were identified constituting 93.37 % of the total oil. The main compounds in major amounts were Isopropyl isothiocyanate (14.03%), (E)-caryophyllene (11.65%), Benzyl isothiocyanate (9.32%), 1,3,8-para-menthatriene(6.61%), Phenyl acetaldehyde (6.55%), Benzane acetonitrile (5.87%), Decanal(5.23%), The compound in trace or in minor amount present in less than 1% were n-hexanol (0.06%), 2-Ethyl-5-methyl tetrahydrofuran-(0.06%), Para-cymene (0.10%), Hex-(3Z)-enyl-butyrate(0.12%), Cembrene (0.16%), Torulosol (0.16%), The oxygenated monoterpene hydrocarbons constituted major portion of the oil (25.40%), followed by sesquiterpene hydrocarbons (23.70%), monoterpene hydrocarbons (7.08%), oxygenated sesquiterpens (4.41%), and oxygenated diterpenes (1.26%), and diterpenes hydrocarbons were in least

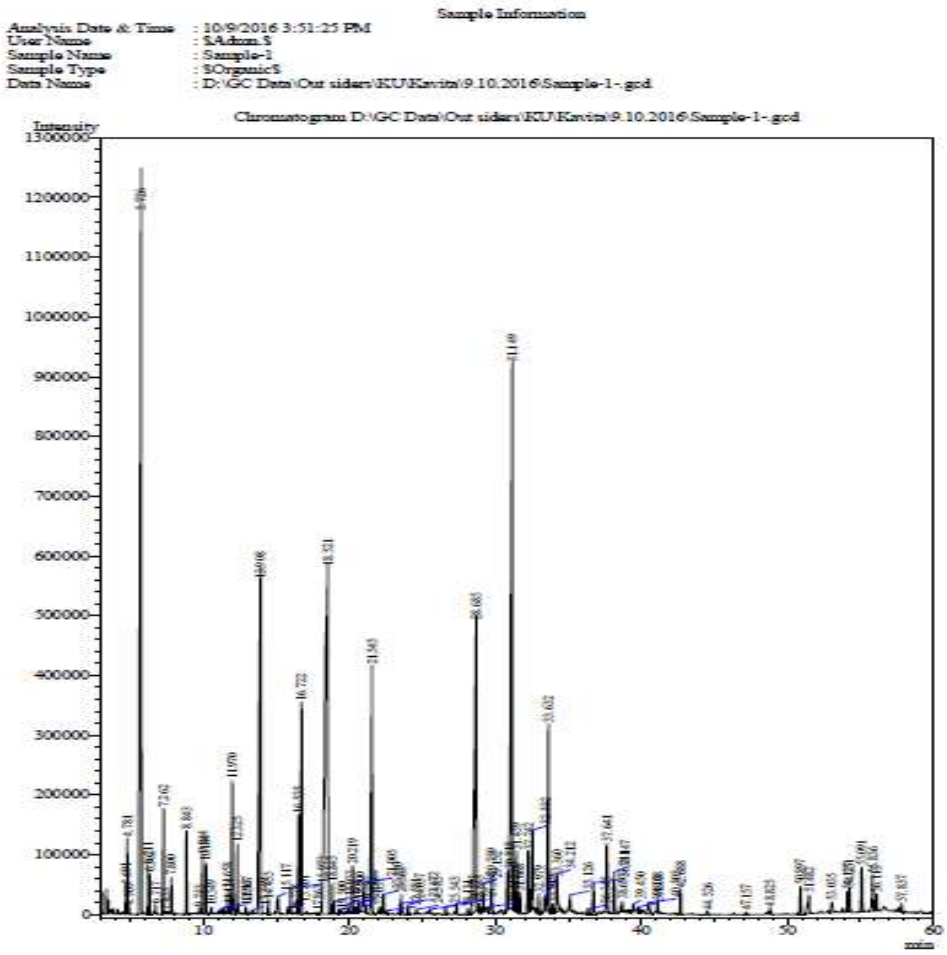
amount (0.16%), so result shows oxygenated monoterpenes were found in the oil as major components while diterpene hydrocarbons as were minor components.

Table1. Essential oil composition of *Excoecaria acerifolia.*

S.N.	Compound	Area %	Mol. formula	Mol. Wt.	RI	Mode of identification
1.	1-Nitro-2-methyl propene	0.48	$C_4H_7NO_2$	101	786	a,b
2.	2-Ethyl-5-methyltetrahydrofuran	0.06	$C_7H_{14}O$	114	810	a,b
3.	Isopropyl isothiocyanate	14.03	C_4H_7NS	101	837	a,b
4.	(2e)-hexenal	0.34	$C_6H_{10}O$	98	845	a,b
5.	(Z)-3-hexenol	0.31	$C_6H_{12}O$	100	853	a,b
6.	N- hexanol	0.06	$C_6H_{14}O$	102	867	a,b
7.	Heptanal	0.41	$C_7H_{14}O$	114	906	a,b
8.	(2e)-heptenal	0.64	$C_7H_{12}O$	112	956	a,b
9.	Benzaldehyde	0.55	C_7H_6O	106	960	a,b
10.	Octanal	2.08	$C_8H_{16}O$	128	990	a,b
11.	N-Butyl isothiocyanate	1.04	C_5H_9NS	115	1001	a,b
12.	Heptadienal	0.27	$C_7H_{10}O$	110	1013	a,b
13.	Para-cymene	0.10	$C_{10}H_{14}$	134	1025	a,b
14.	Phenylacetaldehyde	6.55	C_8H_8O	120	1045	a,b
15.	2E-Octen-1-al	0.22	$C_8H_{14}O$	126	1053	a,b
16.	Cis-Linalool oxide	0.35	$C_{10}H_{18}O_2$	170	1069	a,b
17.	Para-cymenene	0.37	$C_{10}H_{12}$	132	1093	a,b
18.	Linalool	1.91	$C_{10}H_{18}O$	154	1101	a,b
19.	Ho-trienol	3.17	$C_{10}H_{16}O$	152	1102	a,b
20.	1,3,8-para- menthatriene	6.61	$C_{10}H_{14}$	134	1110	a,b
21.	Benzeneacetonitrile	5.87	C_8H_7N	117	1138	a,b
22.	1,4-Dimethyl-4-acetyl-1-cyclohexene	0.53	$C_{10}H_{16}O$	152	1161	a,b
23.	2e-nonenal	0.26	$C_9H_{16}O$	140	1163	a,b
24.	Limonene-4-ol	0.81	$C_{10}H_{16}O$	152	1178	a,b
25.	Hex-(3Z)-enyl butyrate	0.12	$C_{10}H_{18}O_2$	170	1187	a,b
26.	Alpha- terpineol	0.88	$C_{10}H_{18}O$	154	1188	a,b
27.	Decanal	5.23	$C_{10}H_{20}O$	156	1190	a,b
28.	B-Cyclocitral	0.16	$C_{10}H_{16}O$	152	1204	a,b
29.	Myrtenyl acetate	0.37	$C_{12}H_{18}O_2$	194	1234	a,b
30.	Geraniol	0.29	$C_{10}H_{18}O$	154	1255	a,b
31.	(E)-2-decenal	0.17	$C_{10}H_{18}O$	154	1265	a,b
32.	Δ-Elemene	0.24	$C_{15}H_{24}$	204	1335	a,b
33.	Benzyl isothiocyanate	9.32	C_8H_7NS	149	1361	a,b
34.	Alpha- copaene	0.58	$C_{15}H_{24}$	204	1375	a,b
35.	Beta-elemen	0.89	$C_{15}H_{24}$	204	1389	a,b
36.	Alpha- barbatene	0.92	$C_{15}H_{24}$	204	1414	a,b
37.	(E)- caryophyllene	11.65	$C_{15}H_{24}$	204	1424	a,b
38.	Gamma.-elemene	1.40	$C_{15}H_{24}$	204	1431	a,b
39.	Cis- thujopsene	0.52	$C_{15}H_{24}$	204	1433	a,b

40.	Beta- barbatene	1.10	$C_{15}H_{24}$	204	1445	a,b
41.	Alpha humulene	1.75	$C_{15}H_{24}$	204	1454	a,b
42.	4-epi-alpha-acoradiene	0.40	$C_{15}H_{24}$	204	1463	a,b
43.	Germacrene D	3.22	$C_{15}H_{24}$	204	1480	a,b
44.	Gama-amorphene	0.26	$C_{15}H_{24}$	204	1492	a,b
45.	Alpha-selinene	0.59	$C_{15}H_{24}$	204	1494	a,b
46.	Pseudowiddrene	0.16	$C_{15}H_{24}$	204	1503	a,b
47.	Beta-cadinene	0.30	$C_{15}H_{24}$	204	1518	a,b
48.	Caryophyllene oxide	1.53	$C_{15}H_{24}O$	220	1546	a,b
49.	Spathulenol	0.86	$C_{15}H_{24}O$	220	1572	a,b
50.	Humulene epoxide II	0.40	$C_{15}H_{24}O$	220	1613	a,b
51.	Himachalol	0.24	$C_{15}H_{26}O$	222	1650	a,b
52.	Acorenone B	0.41	$C_{15}H_{24}O$	220	1679	a,b
53.	Cembrene	0.16	$C_{20}H_{32}$	272	1937	a,b
54.	N- Hexadecanoic acid	0.97	$C_{16}H_{32}O_2$	256	1977	a, b
55.	Manool	0.42	$C_{20}H_{34}O$	290	2062	a, b
56.	Phytol	0.68	$C_{20}H_{40}O$	296	2114	a, b
57.	Torulosol	0.16	$C_{20}H_{34}O_2$	306	2362	a, b
		93.37				

a=Retention Index (RI), b=MS (GC-MS)



GC-Chromatogram of *Excocaria acerifola*

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