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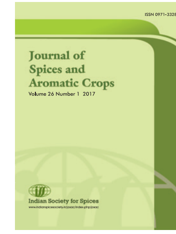

Volatile oil composition of four popular varieties of large cardamom (*Amomum subulatum* Roxb.)

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Abstract

In the present study volatile oil profile of the four popular varities of large cardamom (*Amomum subulatum* Roxb.) namely *Sawney*, *Varlangey*, *Ramla* and *Ramsey* were studied in detail. The capsules contained 58.1- 68.1% seeds and 1.2-2.8% essential oil. By GC-MS analysis 23 constituents of the oil were identified with 1, 8- cineole as the chief component. Major constituents of the oil were α-pinene (1.7-6.5%), β - pinene (4.0-11.1%), 1, 8- cineole (63.2-73.4%) and α- terpineol (5.6-7.6%). The oil contained high level of 1, 8- cineole when compared to small cardamom (*Elettaria cardamom* Maton). α -Terpinyl acetate, the chief component of small cardamom oil was not detected in large cardamom.

Keywords: 1,8-Cineole, essential oil, GC-MS, large cardamom, oleoresin

Large cardamom (*Amomum subulatum* Roxb., Zingiberaceae family, order *Scitamineae*), a cash crop mainly confined to the state of Sikkim in India and Nepal, is now spreading to other states *viz.*, Nagaland, Arunachal, Uttaranchal, Darjeeling and some other parts of North Eastern India, covering an area of about 36,000 ha. The annual production of large cardamom in India varies from 8000 - 8500 MT of which Sikkim alone contributes 3863MT from an area of 26,459 ha (Gudade 2013). It is also cultivated to a lesser extent in Bhutan, Myanmar and China. It is the major cash crop in Sikkim and the second most important source of income and livelihoods (Partap *et al.* 2014). There are 6 cultivars of large cardamom that are specific to altitudes. These are *Seremna* and *Golsey* for low altitude (< 900 m above MSL), *Sawney* and *Ramla* suited to mid altitude (900 - 1500 m above MSL) and *Ramsey* and *Varlangey* specific to high altitude (> 1500 m above MSL).

Large cardamom seeds are arid, bitter and aromatic. It is credited with curative properties in Ayurvedic and Unani systems of medicine (Mukherjee 1972; Singh 1978). The quality of large cardamom depends mainly on colour, size, shape, consistency, texture, and flavor. The flavour and odour of large cardamom is dependent on the composition of aromatic compounds. It is used as a spice as well as in many Ayurvedic preparations. It contains 2-3%

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essential oil and is stomachic, carminative, diuretic and cardiac stimulant, and a good remedy for throat and respiratory trouble. It is used for the treatment of indigestion, vomiting, biliousness, abdominal pains and rectal diseases. Cardio-adaptogenic property against physical stress is also reported by Verma *et al.* (2010). It maintains gastrointestinal, cardiovascular and dental health as well as it provides respiratory relief. Large cardamom oil is used in foods, beverages, mouth fresheners and native medicine. In the present study large cardamom grown in five different locations was evaluated for the essential oil content and its constituents.

Capsules of four cultivars *viz., Ramsey, Ramla, Sawney* and *Varlangey* were collected from different regions of the North Eastern states of India namely Sikkim, Nagaland and West Bengal and one from Myanmar (Burma). Morphological characters of these are given in Table 1. Capsules were harvested at full maturity stage and were dried to the moisture content of 10-12%. Dried large cardamom capsules (20 g per genotype per replication) were crushed and the seeds were separated and weighed. The decorticated seeds were crushed and subjected to hydro-distillation in a Clevenger-type apparatus for 3h and the volatile oil yield was recorded. The oil was dried over anhydrous sodium sulphate and kept in refrigerator until the analysis was carried out. GC analysis of the oil was carried out using a Shimadzu GC-2010 gas chromatograph equipped with FID detector. RTX–5 column (30 m x 0.25 mm, film thickness 0.25 µm) was used. Nitrogen was used as the carrier gas at a flow rate of 1 mL minute^{-1}. The injection port was maintained at 240°C, the detector temperature was 200°C. The oven was programmed as follows: at 65°C for 2 min. and then increased to 155°C @ 3°C/min. The split ratio was 1:50 and 0.2 µL sample was injected. The concentration of each compound was determined by area normalization.

GC-MS analysis was carried out using a Shimadzu GC-2010 gas chromatograph equipped with QP 2010 mass spectrometer and RTX–5 column (30 m x 0.25 mm, film thickness 0.25 µm). Helium was used as the carrier gas at a flow rate of 1 mL minute^{-1}. The injection port was maintained at 240°C, the detector temperature was 200°C. The oven was programmed as indicated above. Ionization voltage was maintained at 70 eV. 0.2 µL sample was injected. The compounds were identified by comparison of retention indices, and by matching the mass spectrum of individual compounds with that of NIST & Wiley library and published literature (Adams 2004).

Aroma and flavor of large cardamom depends on the essential oil, oleoresin and chemical constituents present. Among the samples collected from different locations the percentage

Table 1. Morphological characters of large cardamom cultivars

Cultivars	Characters
Ramsey	Well suited to high altitudes (1515 m above MSL and above) and can be cultivated even in steep slopes. Tillers are maroonish colour and narrow leaves. Capsules are smaller in size varies from 2.0-2.3 cm length and dark pinkish in colour with 25 - 40 seeds.
Ramla	Colour of tillers resembles that of Ramsey but leaves are broad and long. Capsules size varies from 2.0-2.3 cm length and dark pinkish in colour with 30 - 40 seeds.
Sawney	Widely adapted cultivar, which is most suited to medium (975 – 1515 m aMSL) and high (>1515 m aMSL) altitude areas. Leaves are ovate and broad and the colour of tiller is similar to Ramsey. Capsules are bigger and pinkish in colour with 35 - 50 seeds.
Varlangey	Grown in mid and high altitude (>1515 m aMSL) areas. Yield performance is exceptionally high at high altitudes. Leaves are narrow having wavy margins. The productive tiller and spike ratio is relatively high in this cultivar. Capsules are bold in size and pinkish in colour with 50 - 70 seeds.

Table 2. Seed recovery and oil content in the large cardamom cultivars

Characters	Cultivars / Region								
	Sawney			*Varlangey*			*Ramla*		*Ramsey*
	Pangthang (Sikkim)	Nagaland	Myanmar (Burma)	Pangthang (Sikkim)	Kalimpong (West Bengal)	Kalimpong (West Bengal)	Upper Lingzey (East Sikkim)	Kalimpong (West Bengal)	Upper Lingzey (East Sikkim)
Seed recovery (%)	55.9	68.0	62.4	58.3	66.5	68.1	59.1	65.3	65.2
Oil on capsule weight basis (%)	1.5	2.4	2.4	1.2	1.8	2.4	1.2	2.7	2.8

seed recovery varied from 55.07-68.10%. The highest seed recovery was obtained in *Varlangey* from Kalimpong (68.10%) which was followed by *Sawney* collected from Nagaland (68.0). *Sawney* from Sikkim recorded the lowest seed recovery (55.9%) (Table 2).

In large cardamom capsules essential oil yield varied from 1.2-2.8% with the highest recorded in *Ramsey* collected from Upper Lingzey, East Sikkim, which was followed by *Ramla* from Kalimpong (2.7%). In case of the *Sawney* collected from different regions, samples collected from Nagaland and Myanmar had higher oil yield (2.4%) compared to that from Pangthang (Sikkim, 1.5%). In the case of *Varlangey*, samples from Kalimpong recorded the higher oil content (2.4%) in comparison with that from Sikkim. In *Ramla* also samples from Kalimpong recorded the higher oil yield (2.7%) compared to that from Sikkim (Table 2).

GC-MS profile of large cardamom oil is shown in Fig. 2. Chemical constituents identified, by GC and GC-MS analyses are given in Table 3. Twenty three compounds contributing 92.9-98.2% of the oil were identified by GC-MS analysis of which monoterpene hydrocarbons contributed 7.87-21.0% and oxygenated monoterpens 75.5-84.0%. The chief component of the oil, 1, 8-cineole varied between 63.2-73.4%. The samples from Sikkim contained relatively lower level of 1, 8 cineole. α-pinene (1.7-6.5%), β-pinene (4.0-11.1%) and α-terpeniol (5.6-7.6%) were the other major compounds identified. Sabinene (0.4-2.2%), myrcene, γ-terpinene, 4- terpineol, nerolidol and sabinene hydrate were the minor components which contributed to the flavour of oil.

Among the cultivars from different agro-climatic zones *Ramla* from Kalimpong recorded the highest content of 1, 8-cineole followed by *Varlangey* cultivar from Kalimpong (73.0%). 1, 8 cineole is a remarkable chemical component offering strong therapeutic properties that has been well researched. It has strong healing potential. It has anti-inflammatory (Santos & Rao 2000; Juergens *et al.* 2003; Juergens *et al.* 2004; Santos *et al.* 2004), antibacterial,

Table 3. Chemical constituents of large cardamom oil

Adams Index	Compounds	Composition of essential oil (%) *Sawney* Pangthang (Sikkim)	*Sawney* Nagaland	*Sawney* Myanmar (Burma)	*Varlangey* Pangthang (Sikkim)	*Varlangey* Kalimpong (West Bengal)	*Varlangey* Kalimpong (West Bengal)	*Ramla* Upper Lingzey (East Sikkim)	*Ramla* Kalimpong (West Bengal)	*Ramsey* Upper Lingzey (East Sikkim)
0929	α-Thujene	0.4	0.3	0.4	0.5	0.1	0.3	0.5	0.3	0.3
0932	α-Pinene	4.7	3.8	4.0	6.5	1.7	3.8	5.8	4.1	4.1
0969	Sabinene	-	-	1.6	0.4	0.9	1.3	-	1.3	2.2
0974	β-Pinene	8.6	7.1	5.4	11.1	4.0	5.3	10.7	5.3	6.3
0988	Myrcene	-	1.1	1.9	0.7	0.7	1.41	1.1	1.6	1.4
1002	α-Phellan-drene	-	0.3	-	-	-	-	-	-	-
1014	α -Terpinene	0.4	-	0.3	0.5	0.1	0.34	0.4	0.4	-
1025	β -Phellan-drene	-	1.5	-	-	-	-	-	-	-
1026	1,8 – Cineole	68.8	72.6	70.1	63.5	73.0	71.0	63.2	73.4	69.5
1032	β -Ocimene	-	-	0.6	-	-	-	-	-	-
1054	γ-Terpinene	1.0	0.6	1.2	1.1	0.3	1.1	1.2	1.0	0.3
1065	Sabinene hydrate	-	0.9	-	-	1.9	-	-	-	2.5
1086	α -Terpinolene	0.3	0.1	0.3	0.3	0.1	0.3	0.3	0.2	0.1
1135	Trans pino-carveol	-	-	-	0.3	-	-	0.4	-	-

Contd.

Adams Index	Compounds	Composition of essential oil (%)								
		Sawney			*Varlangey*			*Ramla*		*Ramsey*
		Pangthang (Sikkim)	Nagaland	Myanmar (Burma)	Pangthang (Sikkim)	Kalimpong (West Bengal)	Kalimpong (West Bengal)	Upper Lingzey (East Sikkim)	Kalimpong (West Bengal)	Upper Lingzey (East Sikkim)
1140	β -Terpineol	-	-	-	-	-	-	-	-	0.8
1160	pinocarvone	-	-	-	-	-	-	0.1	-	-
1174	4-Terpineol	-	1.8	3.2	3.2	-	3.0	3.4	2.7	0.9
1182	Cis- pino- carveol	-	0.2	-	-	1.1	-	-	-	-
1186	α-Terpeneol	6.5	6.2	7.4	5.6	6.5	7.6	5.8	6.7	6.7
1194	Myrtenol	0.1	-	-	0.3	-	-	-	-	-
1437	α -Guaiene	-	0.1	-	-	-	-	-	-	-
1489	β-Selinene	-	-	-	-	-	-	0.1	-	-
1561	Nerolidol	2.1	1.4	1.3	2.6	1.4	1.5	3.0	1.2	-
	Total %	92.9	98	97.7	96.6	91.8	96.95	96	98.2	95.1
	Monoterpene hyrdocarbns	15.4	14.78	15.62	21.06	7.87	13.8	19.99	14.23	14.66
	Oxygenated monoterpenes	77.5	82.19	81.9	75.5	82.0	83.1	76.0	84.0	77.6

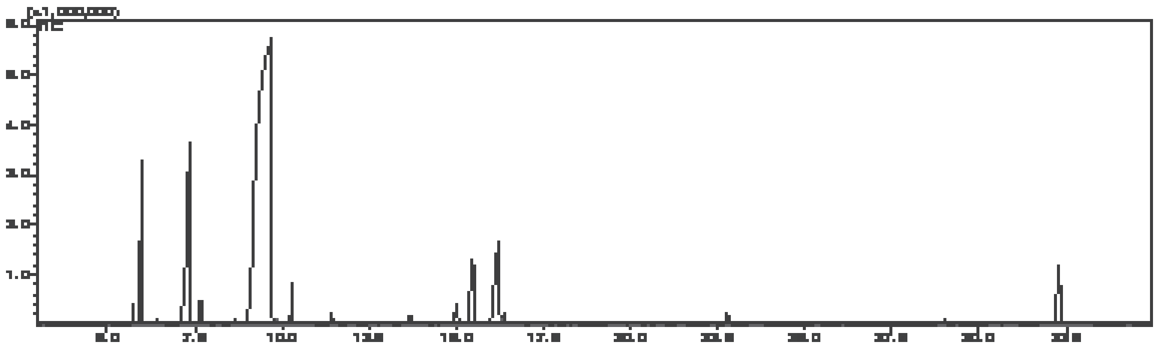

Fig. 2. GC-MS Profile of large cardamom oil

antioxidant, antispasmodic (Nascimento *et al.* 2009; Coelho-de-Souza *et al.* 2005), antiviral (Astani *et al.* 2010) and hypotensive properties (Lahlou *et al.* 2002). *Ramla* and *Varalangi* from Kalimpong (West Bengal), *Sawney* from Nagaland and Burma had high cineole content where as the cultivars from Sikkim recorded low 1,8-cineole and high α- and β- pinenes. Not much variation was observed with respect to the composition of other constituents. As observed by several workers (Gurudatt *et al*. 1996) α-terpinyl acetate the major constituent of small cardamom oil, was not detected in our samples. β-Terpinyl acetate which has a pleasant fruity smell, contribute to the superior quality of small cardamom. However, Bhandari *et al*. (2013) reported 3.3% α-terpinyl acetate, in the large cardamom oil of cv. *Sawney* from Uttarakhand. Similarly Joshi *et al.* (2013) also reported the presence of α-terpinyl acetate in the large cardamom collected from Himachal Pradesh.

Several constituents of the large cardamom oil are reported to have medicinal properties. 1,8 – cineole, terpeniol, sabinene and α-pinene act as tonic for the heart and liver and are good appetizers, and promote the elimination of bile and help to reduce congestion of the liver (Chempakam & Sindhu 2008). α-Pinene has anti-inflammatory and broad-spectrum antibiotic (Nissen *et al.* 2010) activities. Among the cultivars studied variations exist in essential oil, oleoresin content and chemical constituents which is due to the varietal characters and variations within the cultivar from different regions are mainly due to the effect of environmental factors. The present study highlights the importance of selecting region-specific cultivars for better profits.

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