Journal of Essential Oil Bearing Plants

Publication details, including instructions for authors and subscription information:

<http://www.tandfonline.com/loi/teop20>

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Published online: 16 Apr 2015.

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To cite this article: P. Vyshali, K.J. Thara Saraswathi & G.R. Mallavarapu (2015) Chemical Composition of the Essential Oils of *Cymbopogon citratus* (DC.) Stapf Grown in Three Locations in South India, *Journal of Essential Oil Bearing Plants*, 18:1, 230-235, DOI: [10.1080/0972060X.2014.935075](https://doi.org/10.1080/0972060X.2014.935075)

To link to this article: <http://dx.doi.org/10.1080/0972060X.2014.935075>

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Chemical Composition of the Essential Oils of *Cymbopogon citratus* (DC.) Stapf Grown in Three Locations in South India

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Received 07 April 2013; accepted in revised form 21 November 2013

Abstract: Essential oils of *Cymbopogon citratus* (DC.) Stapf grown in three different locations in South India were analysed by GC and GC-MS. The main constituents of the oils from Bangalore and Puttur were identified as β -myrcene, neral, geranial and geraniol. The oil from Nadugani was found to contain the major constituents' neral, geranial, geraniol and geranyl acetate. The present study revealed the presence of six oxygenated monoterpenes and sesquiterpenes which were hitherto not reported in the oil of *C. citratus*.

Key words: *Cymbopogon citratus* (DC.) Stapf, Poaceae, essential oil composition, β -myrcene, neral, geranial, geraniol, geranyl acetate, monoterpenes, sesquiterpenes.

Introduction

Cymbopogon citratus (DC.) Stapf (Syn. *Andropogon citratus* DC.) popularly known as West Indian lemongrass belonging to the family Poaceae is a native of South West Asia and is grown in several countries particularly West Indies, China, Indonesia, Brazil, Congo, Republic of Malagasy, Sri Lanka, Zambia and other countries ^{1,2}. The other two types of lemongrass viz. East Indian lemongrass (*C. flexuosus*) and the North Indian lemongrass (*C. pendulus*) as well as the hybrid CKP-25 (*C. khasianus* and *C. pendulus*) are commercially cultivated in India for the production of lemongrass oil. *C. flexuosus* oil and the oils of the improved varieties Pragathi, Praman, Cauveri, Krishna derived from *C. flexuosus* and the oil of CKP-25 are considered superior in quality because of their solubility in 70 % alcohol and high content of citral (neral + geranial) ². West Indian lemongrass oil is considered inferior because the oil has low solubility in 70 % alcohol

and also due to low citral content. Also the oil has the tendency to polymerise because of high content (5-25 %) of the monoterpene β -myrcene in the oil, thus deteriorating the quality of the oil ¹.

This oil has been the subject of several studies by many researchers from different countries ³⁻¹⁹. According to a recent review a total of 158 compounds were identified in this oil by several research groups from different countries ². The identified compounds represent monoterpenes, oxygenated monoterpenes, sesquiterpenes, oxygenated sesquiterpenes, phenyl propanoids, aliphatic compounds and nitrogenous compounds. Most of the studies on this oil reported the main constituent of the oil as citral (neral and geranial) except the oil from Ethiopia which was reported to contain geraniol (40 %), citral (13 %) and α -oxobisabolene (12 %) as the major compounds ¹.

A recent study of the oil of this plant from Delhi (India) reported the presence of 1,8-cineole, nerol, neral and geraniol as the major components ¹⁹.

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There are only three reports on the oil composition of this plant from India¹⁷⁻¹⁹. As there is wide variation in the identification of the minor components of this oil from different locations and countries, we have investigated *C. citratus* grown in three locations in South India with the objective to explore the possibilities for commercial application of this oil based on their chemical composition. We report here the results of our study on the composition of the oils from Bangalore, Puttur and Nadugani in South India.

Experimental plant material

Plants of *C. citratus* grown in Bangalore nursery, in Puttur (South Canara District, Karnataka) and Nadugani (Nilgiri district, Tamil Nadu) were collected and the shade-dried leaf materials from the three places were used in the present study. Voucher specimens were deposited in the herbarium maintained by Biotechnology Department of Bangalore University.

Isolation of essential oils

The shade-dried leaf materials from the three locations (200 g. each) were separately hydro-distilled in a Clevenger-like apparatus for 3 hrs. The pale yellow essential oils with strong, fresh citrus odor obtained were collected and dried over anhydrous Na₂SO₄ and stored in a refrigerator until analyzed. The oil contents of the materials were 0.8 % (Bangalore), 0.7 % (both Puttur and Nadugani) (on dry wt basis).

Gas chromatographic and Gas chromatographic-Mass spectral analysis

GC analysis of the oil samples was performed on an Agilent Technologies gas chromatograph Model 6890N equipped with dual FID. A CPSil8CB column (30 m X 0.25 mm X 0.25 µm film thickness) coated with dimethylpolysiloxane with 5 % diphenyl as the stationary phase. Helium was used as the carrier gas at a flow rate of 1 ml per min. (constant flow). Temperature programming was done from 50°C (2 min.) 280°C at 10°C per min. Injector and detector temperature were maintained at 250°C and 280°C respectively. Samples of 1 µl dissolved in hexane were injected using a split ratio of 10:1. GC-MS was done using

the above GC interfaced with mass selective detector (Agilent technologies 5973). The same stationary phase as GC was used. Helium was used as the carrier gas. GC conditions same as above. Mass spectra were recorded in the EI mode at 70 eV in the m/z range of 30-450.

Component identification was done by comparison of linear retention indices of GC peaks obtained using alkanes (C8-C25) with those of compounds reported in literature²⁰ and by comparison of the mass spectra of the peaks with those of compounds from literature²⁰ and those stored in NIST library. Peak area percentages were computed from GC peak areas without using correction factors.

Results and discussion

Hydrodistillation of the shade dried leaves of *C. citratus* collected from Bangalore, Puttur and Nadugani afforded pale yellow essential oils having strong fresh grassy citrus odor. The oil content of the leaves from the three locations was found to be in the range 0.7-0.8 %. GC and GC-MS analysis of the oils from the three places enabled the identification of compounds which are listed in Table 1. The identified compounds include monoterpenes, oxygenated monoterpenes, sesquiterpenes, oxygenated sesquiterpenes, aliphatic compounds and phenylpropanoids. Table 1 shows that there are similarities as well as differences in the composition of the oils of the plant from the three places. Oxygenated monoterpenes dominated the oils. Citral (neral + geranial) is main component of the oils of three places and therefore the oils from the three locations belong to citral chemotype. β-Myrcene was found as a major component of the oils from Bangalore and Puttur whereas the oil from Nagudani was found to contain geraniol and geranyl acetate as major components. The oil from Bangalore contained high concentration of neral and geranial than the other two oils from Puttur and Nadugani. The oil from Nadugani had higher concentration of geraniol and geranyl acetate than the other two oils from Bangalore and Puttur. The monoterpene β-myrcene which is normally found in a concentration of 5-25 % in West Indian lemongrass oil was found as a minor component in the oil from

Table 1. Chemical compositions of the essential oils of *Cymbopogon citratus* leaf collected from Bangalore, Puttur and Nadugani

RI	Compound	Percentage		
		Bangalore	Puttur	Nadugani
937	α -Pinene	tr	tr	tr
986	6-Methyl 5-hepten-2-one	0.6	0.5	0.7
990	β -Myrcene	8.8	6.9	0.2
1000	Octanal	nd	tr	0.2
1026	p-Cymene	0.1	0.1	0.1
1030	Limonene	0.5	tr	1.4
1038	(Z)- β -Ocimene	0.1	0.2	0.4
1048	(E)- β -Ocimene	tr	0.1	0.3
1053	2,7-Dimethyloctadiene	nd	0.1	0.1
1069	4-Nonanone	nd	nd	0.1
1081	6-Camphenolone	0.7	nd	nd
1088	6,7-Myrcene epoxide	0.3	0.8	0.1
1090	Terpinolene	nd	nd	0.1
1092	Perillene	nd	0.1	0.1
1098	Linalool	1.3	1.3	1.2
1103	2,2-Dimethyl 3,4-octadien-1-al	0.4	0.1	0.3
1111	epi-Photocitral B	0.1	0.6	tr
1143	<i>exo</i> -Isocitral	1.0	0.7	0.7
1151	Citronellal	0.3	0.6	1.2
1165	(Z)-Isocitral	0.9	1.9	1.9
1174	Rosefuran epoxide	0.1	0.1	0.3
1184	(E)-Isocitral	1.4	2.0	2.8
1204	Decanal	0.1	0.5	0.6
1229	Citronellol + Nerol	0.5	0.2	0.2
1259	Neral	30.4	24.9	23.8
1260	Geraniol	2.2	3.1	17.1
1280	Geranial	41.8	28.9	23.3
1298	2-Undecanone	0.1	0.4	0.5
1305	Geranyl formate	0.1	0.7	0.2
1344	6,7-Epoxyneral ^{ti} + 2,4-Octanediol ^{ti}	0.6	2.1	0.4
1358	Neric acid	0.3	0.8	0.6
1362	Citronellyl acetate	0.1	nd	0.2
1377	6,7-Epoxygeranial ^{ti}	0.3	2.7	0.4
1385	Geranyl acetate	0.4	1.2	5.8
1387	Geranic acid	0.3	1.1	0.4
1392	β -Elemene	0.1	0.9	0.3
1419	(E)-Caryophyllene	tr	0.5	1.7
1436	<i>trans</i> - α -Bergamotene	tr	0.3	0.2
1442	α -Guaiene	0.1	nd	0.2
1446	(E)-Isoeugenol	nd	nd	0.4
1456	α -Humulene	tr	nd	0.4
1478	γ -Murolene	tr	0.2	0.1

table 1. (continued).

RI	Compound	Percentage		
		Bangalore	Puttur	Nadugani
1482	Germacrene D	tr	0.2	0.3
1497	Tridecan-2-one	0.3	nd	0.1
1500	α -Selinene	0.1	0.3	0.1
1513	γ -Cadinene	0.4	0.2	0.3
1525	δ -Cadinene	0.4	0.2	0.4
1552	Elemol	nd	0.1	0.5
1572	Germacren-D-4-ol	tr	0.2	0.4
1583	Caryophyllene oxide	0.3	0.4	1.4
1605	Humulene epoxide II	0.1	0.5	0.3
1617	Selin-6-en-4 α -ol	0.1	0.6	0.7
1638	τ -Muurolol	nd	0.3	0.3
1643	α -Muurolol	0.1	nd	0.4
1945	Hexadecanoic acid (Palmitic acid)	0.1	nd	nd
2100	Heneicosane	0.1	nd	nd
2200	Docosane	0.1	nd	nd
	Geranyl geranate ^{ti}	0.1	0.4	0.1
	Monoterpenes	9.5	7.4	2.6
	Oxygenated monoterpenes	84.0		
	Sesquiterpenes and oxygenated sesquiterpenes			

RI = Retention Index on DB-5 column;

nd = not detected;

tr = trace (>0.05%).

ti = tentative identification

Nadugani. The oils from Bangalore and Puttur had this compound in a concentration of 8.3 % and 6.3 % respectively. The oils from the three places also contained sesquiterpenes and oxygenated sesquiterpenes. The fatty acid palmitic acid and the alkanes heneicosane which were detected in the oil from Bangalore were not found in the oils from Puttur and Nadugani. *Exo*-isocitral was reported earlier in the oil of *C. citratus* from Colombia as a minor constituent. Identification of the compounds 2,4-octanediol, 6,7-epoxyneral and 6,7-epoxygeranial was tentative based on their mass spectral data. The monoterpene aldehydes (Z)-isocitral and (E)-isocitral, 6,7-epoxyneral and 6,7-epoxygeranial were not found earlier in *C. citratus* oil and are being reported for the first time in these oils. The mass spectral data of these compounds are presented in Table 2. The phenylpropanoid (E)-isoeugenol was detected only

in the oil from Nadugani and not in the oils from Bangalore and Puttur. The oils from Puttur and Nadugani contained relatively high concentration of sesquiterpenes and oxygenated sesquiterpenes (5.3-7.6 %) than the oil from Bangalore which had these compounds in a concentration of only 1.4 %. The variation in the essential oil composition can be attributed to the phytogeographic and climatic factors as discussed in earlier reports ^{21, 22}.

West Indian lemongrass oil was reported to find application in cosmetic and perfumery industries and in the preparation of herbal tea. The main component of the oil citral was used as a starting material for the preparation of ionones which are used in violet perfumery. β -Ionone was used as a raw material for the production of vitamin A. This was reported to possess several biological activities such as antibacterial and antifungal activities. Besides, citral from lemongrass oil was reported

Table 2. Mass spectral data of the tentatively identified compounds and the compounds detected for the first time in *C. citratus* essential oil

RI	Compound	Mass fragments (m/z)
1144	<i>exo</i> -Isocitral	41(52), 53(10), 55(5), 63(2), 65(5), 67(10), 69(100), 77(5), 79(5), 81(10), 91(8), 93(5), 109(40), 119(3), 121(2), 123(2), 126(1), 137 (2), 152(5).
1165	(Z)-Isocitral	41(60), 43(20), 53(25), 55(30), 59(5), 63(3), 63(10), 67(85), 69(40), 77(35), 79(40), 81(90), 91(40), 93(35), 94(90), 105(10), 109(100), 119(30), 121(3), 123(5), 134(3), 137(10), 152(5).
1184	(E)-Isocitral	41(50), 43(25), 53(20), 55(25), 65(10), 67(70), 69(25), 77(20), 79(28), 81(100), 91(25), 93(23), 94(80), 105(5), 107(8), 109(80), 119(15), 121(5), 123(12), 134(5), 137(8), 152(5).
1344	6,7-Epoxyneral ^{ti} + 2,4-octanediol ^{ti}	41(30), 43(20), 53(5), 55(8), 57(7), 59(40), 65(5), 67(15), 71(7), 77(6), 79(15), 81(20), 82(100), 85(14), 91(5), 93(3), 95(30), 97(20), 107(2), 110(10), 125(3), 135(1), 139(1), 153(2), 168(3).
1377	6,7-Epoxygeranial	41(52), 43(25), 53(10), 55(25), 57(30), 59(85), 67(20), 69(23), 71(20), 77(10), 79(10), 81(100), 83(15), 85(90), 91(5), 93(4), 95(20), 97(50), 100(2), 109(8), 125(10), 139(5), 153(4), 168(2).

ti = tentative identification

to be active against P388 leukemic cells ²³. Out of the three oils of the present study, the oil from Nadugani with low citral content and moderate amounts of geraniol and geranyl acetate may find application in food flavours. The germplasm of this plant has the potential for commercial cultivation and production of the essential oil. Development of improved varieties with high oil

content and high citral content may help in commercial exploitation of this plant.

Acknowledgement

The authors are beholden to the Mass spectra Centre of Indian Institute of Chemical Technology, Hyderabad for the GC and GC-MS analyses of the oils.

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