INT J CURR SCI 2013, 7: E 42-47

RESEARCH ARTICLE

ISSN 2250-1770

Study on the essential oil of aerial and sub-aerial parts of *Cymbopogon flexuosus* (Nees ex Steud) Wats.

Vinutha M and K.J. Thara Saraswathi

Department of Microbiology and Biotechnology, Bangalore University

Bangalore-560 056, Karnataka, India

Corresponding author: dr.tharabiotech@gmail.com; vinutha_gowda@yahoo.co.in

Abstract

Cymbopogon flexuosus (Nees ex Steud) Wats commonly known as, East Indian lemongrass a widely grown essential oil plant in the world belongs to the family Poaceae and comprise of 140 species worldwide, found abundantly in tropics and sub-tropic regions of Asia, Africa and America. In India, 45 species are recorded of which the economic importance is *C.winterianus, C.flexuosus, C.martinii, C.nardus, C.citratus, C.pendulus, C.jwarancusa* and *C.khasianus.* Aerial and sub-aerial parts of *C. flexuosus* collected from Himavath Gopala hills, Karnataka, India, were subjected to hydrodistillation for extraction of essential oil. GC and GC-MS analysis were performed to know the chemical composition of the oil. Among the 39 compounds identified in aerial parts of the plant the major compounds were citral (64.98%), 1,7-octadien-3-ol (10.97%), dimethyl oxatricyclo nonanone (9.44%), nerol (2.85%), verbenol (1.77%) and caryophyllene oxide (0.71%). In sub-aerial parts of the plant 33 compounds were identified. The analysis of sub-aerial parts showed a different chemical profile compared to aerial part and possessed citral as the major compound of upto 30.47%. Other compounds in sub-aerial part are Eudesmol (17.82%), Elemol (14.16%), dihydro isopropyl methyl azulene (11.08%), ë-cadinene (1.88%), junipene (1.36%), hydroxyalloaromadendrene, juniper camphor (1.12%) and elemene (1.04%).

Keywords: *Cymbopogon flexuosus*, essential oil, GC-MS, citral

Received: 28th January; Revised: 26th February; Accepted: 14th April;

Introduction

Cymbopogon flexuosus (Nees ex Steud) Wats is a perennial aromatic cum medicinal herb native to Indian subcontinent and belongs to Poaceae (Hussain et al., 1988). It is commonly known as Cochin, East Indian / Malabar lemongrass (Adinarayana et al., 2012). It is a tufted grass of about 2m height. The leaves are linear and lanceolate, inflorescence highly branched, very large with terminal drooping panicle bearing spikes on tertiary branches. The plant is grown in all types of soil from light sandy soil to upland laterites. It flourishes well in drained sandy loams of pH 7.5. Essential oil percentage in leaves of *C.flexuosus* varies from 0.2 to 0.4% (Joy et al., 2006). In different cultivars of *C.flexuosus* the oil yield varied from 0.7 to 1.0%. The essential oil of *C.flexuosus* contains a mixture of complex volatile constituents utilized in flavour, fragrance, food, perfumery, cosmetics, toiletries, fine chemicals, pharmaceutical industries and in aroma therapy (Parikh and Desai, 2011). They are used as such or in diluted forms in the aromatherapy sector (Curtis, 1996). Bioactivity studies have shown that lemongrass oil possesses anticancer (Sharma et al., 2009), analgesic, anti-inflammatory (Chandrashekar and Prasanna, 2010), antibacterial (Fazrullah Innsan et al., 2011) and antiviral (Chao et al., 2000). The important constituents in the essential oil were, geranial (25-53%), neral (20-45%), limonene (≤0.1%-1.8%), β-caryophyllene (≤0.1%-0.9%) and geranyl acetate (0.1%-0.8%) (Padalia et al., 2011). Citral is a mixture of acyclic monoterpene aldehydes: neral (cis-citral) and geranial (trans-citral) (Rauber et al., 2005). It is the active compound in essential oil of lemongrass and determine the quality of the oil. Higher the citral content, purer the oil (Schaneberg and Khan, 2002). Citral, the chief constituent in oil of lemongrass is the starting material in the production of ionones which are required in the synthesis of several aromatic compounds extensively used in soaps, cosmetics, perfumes and in the production of vitamin-A (Kulkarni, 1994).

Citral and geranyl acetate have extensive uses in pharmaceutical, flavouring, essence and many synthetic preparations (Aniruddha Sarma et al., 2011). The essential oil of *C.flexuosus* var. *flexuosus* exhibits good solubility in

alcohol, hence it is considered as superior in quality (Guenther, 1950). The chemical composition of the volatile oil obtained from the leaf of lemongrass is well studied (Sarma et al., 1998; Nath et al., 2002). The essential oil from the roots of *C. densiflorus* were found to be rich in citral (37%) (Loumouamou et al., 2010). In the rhizome of *C. citratus* the essential oil was rich in selina-6-en-4-ol (27.08%) followed by citral (15%) (Andrade et al., 2009). However the study on the essential oil from rhizome/ sub aerial region has not been studied so far in *C. flexuosus*. Hence, the present work is focussed on studying the sub aerial region of *C. flexuosus* for its essential oil content and composition in comparison with the aerial part.

Materials and Methods

Collection and maintenance of plant

The wild species of *C. flexuosus* were collected from Himavath Gopala hills, Karnataka and maintained in the departmental garden, Bangalore University, Bangalore. The plants collected were identified by Regional Research Institute (RRI), Bangalore, Karnataka. The cytological investigations were carried out. The essential oil was extracted from four month old plants.

Extraction of essential oil: Aerial part (leaves, shoots)

The herbage was cut into small pieces of approximately 25 mm length and air dried at room temperature for 3 days. The fresh and dry weight of the herbage before drying and after drying was noted down. The herbage was subjected for extraction of essential oil.

Sub-aerial part (rhizome, roots)

The sub-aerial parts of the plant was thoroughly washed to remove the soil particles, cut into small pieces of 25 mm length and air dried at room temperature for 5 days. The fresh and dry weight of the rhizome, before and after drying was noted. The extraction of sub aerial part was carried out. The essential oil was extracted through hydrodistillation the most common method for volatile oil extraction (Doina Atofani et al., 2010) using Clevenger's apparatus (Clevenger, 1928). This method is described in the European Pharmacopoeia 2005 (ÖZEK et al., 2010).

Analysis of essential oil: Quantitative analysis

The percentage composition of essential oil obtained from aerial and sub aerial regions were calculated by using the formula, essential oil content (V/W) = a x100 / b

Where a = volume of the oil in ml collected through hydrodistillation

b = weight in grams of the sample taken.

Qualitative analysis

GC method

The GC analysis was performed on a Thermo GC-trace ultra ver: 5.0, equipped with a split/splitness injector (260^0C, split ratio 1:10) using DB-5 capillary standard non-polar column (30 mx0.25 mm, df: 0.25 µm). The temperature program was 70^0c (6 min) rising to 260^0C at a rate of 6^0C/min. Injector and detector temperature was 260^0C. Helium was used as carrier gas at a flow rate 1.0 ml/min.

GCMS method

The GC/MS analysis was performed on a Thermo GC-trace ultra ver: 5.0, Thermo MS DSQ II using DB 5-MS Capillary Standard Non-Polar Column (30 mts x 0.25 mm x 0.25 µm). The temperature program was 70^0C (6 min) rising to 260^0C at a rate of 6^0C/min. Injector and detector temperature was 260^0C. Helium was used as carrier gas at a flow rate 1.0 ml/min. Identification of the compounds was carried out by comparison of the mass spectral fragmentation patterns with those stored in MS database (National Institute of Standards and Technology).

Results

The cytological studies indicate that the plant belongs to the tetraploid race.

Quantitative analysis

Hydrodistillation of aerial and sub aerial parts of the wild species of *C. flexuosus* yielded 1.27 and 0.32% (V/W) of essential oil respectively. The essential oil of aerial part showed deep yellow colour compared to sub aerial part which showed light yellow colour.

Qualitative analysis of essential oil

The composition of essential oil from aerial part of *C.flexuosus* is given in Table 1. The essential oil showed rich geranial content with 35.13% and 29.85% neral. The other major constituents present were 1,7-octadien-3-ol (10.97%), dimethyl oxatricyclo nonanone (9.44%), nerol (2.85%), verbenol (1.77%) and caryophyllene oxide (0.71%), ς- cadinene (0.45%), geranyl acetate (0.44%), trans caryophyllene (0.34%), caryophyllene (0.33%),

Table 1. Chemical composition of essential oil from aerial region

Compound Name	Peak Area (%)
6-Methyl-5- hepten-2-one	0.05%
Linalool	0.16%
α-pinene oxide	0.02%
Cis-Limonene oxide	0.02%
Z-citral	29.85%
E-citral	35.13%
Geranyl acetate	0.44%
Trans-caryophyllene	0.34%
Caryophyllene	0.33%
ς – cadinene	0.45%
Caryophyllene oxide	0.71%
Farnesol	0.01%
Nerol	2.85%
1,7-octadien-3-ol	10.97%
5-Acetoxy-3,4-dimethyl-2-cyclopenten-1-one	0.16%
(2-acetylpyridine-S-Methyldithiocarbazate)dimethyl alumium	0.05%
4- Nonanone	0.11%
3-Oxatricyclo[4.1.1.0(2,4)]octane, 2,7,7-trimethyl	0.02%
5`-5 Dihydroxy-1-1'-bicyclo octylidene methanone dicyclopropyl	0.51%
7-oxabicyclo[4.1.0] heptane, 1-methyl-4 (1-methyl ethenyl)	0.96%
Verbenol-trans	0.96%
Verbenol	1.77%
Trans-carveol	0.07%
(E)-8-acetyloxy-3,4-epoxy-2 (2,4-hexadinylidine)-1,6 dioxaspiro(4,5) decane	0.02%
Fenchone	0.01%
(1R,4S,6R,7S)-1,7-Dimethyl 9-oxatricyclo (4.3.0.0(4,7)) nonanone	9.44%
2-Undecanone	0.03%
Ethanol, 2- 99,12-octadecadienyloxy)-(Z,Z)	0.03%
Neryl acetate	0.22%
Aromadendrene	0.01%
2-ethyl-3-hydroxy-3(methoxymethyl)isoindolinone	0.08%
3-Methoxyphenanthro (3,4-b)(1)benzothiophene 11-oxatetracyclo[4.2.2.2(2,5).1(7,10)]	0.03%
Undec-3-ene-9-methoxy-9-methyl	0.03%
12-oxabicyclo(9.1.0)dodeca-3,7-diene1,5,5,8-tetramethyl[1R-(1R,3E,7E,11R)]	0.03%
(E)-1-phenyl-2(triethylsilyl)ethylene	0.01%
3,7,11,15-tetramethyl-2-hexadecen-1-ol	0.05%
Neoisolongifolene,8-bromo	0.05%
[(1rs.2rs)-2-methyl-2-(4'-methyl-3'penteyl)cyclopropane] carbaldehyde	0.02%
Uvidin A	0.02%
Total	96.02

Table 2. Chemical composition of essential oil from sub aerial region

Compound Name	Peak Area (%)
Elemene	1.04%
Juniper camphor	1.12
ς – terpinene	0.02
Elemol	14.16
Z-citral	8.12
E-citral	22.35
á-Eudesmol	17.82
ë-cadinene	1.88
(E)-2,3-Epoxyoctan-1-ol	0.08
1-[2H3] Methoxyindole	0.08
9-oxabicyclo[6.1.0]non-6-en-2-one	0.16
2-Heptanone,6-methyl	0.06
á-Elemene	1.04
Junipene	1.36
Adrenalone	0.02
Cyclohexanone,5-methyl-2(1-methylethylidene)	0.02
Pulegone	0.22
trans(3R,8aR)-3-8a-dihydro-3-isopropyl-8a-methylazulene-1(2H)one	11.08
(-)-(10R)-Hydroxyalloaromadendrane	1.12
2,4,4-Trimethyl-3(1-hydroxy-2-propenyl-2-cyclohexenone)	0.20
Herquinone	0.04
(3-Fluoro-5-bromophenoxy)amine	0.04
2-(1-cyclopent-1-enyl-1-methylethyl)-cyclopentanone	0.04
Bicyclo[3.3.1]nonan-2-one,9-hydroxy-,syn	0.08
4H-1,4-Epoxy-4a,7-methanonaphthalene,1,5,6,7,8,8a-hexahydro-.(1à, 4à, 4aà,7à,7aà)	0.68
9Z)-se-phenyl-9-cyclohexenylon-5-ensclenoate	0.32
δ-cadinene	1.88
Bicyclo[4.4.0]dec.2-ene-4-ol,2-methyl-9-(prop-1-en-3-ol-2-yl)	0.02
1,2,5,6-tetramethylbiclclo[3.3.0]octan-3-a-ol	0.26
2-Naphthalenemethanol,1,2,3,4,4a,5,6,8a-octahydro- à, à,4à, 8-tetramethyl-(2à,4aà,8aà)	1.20
2-Naphthalenemethanol, decahydro-a, à, 4a-trimethyl-8-methylene-[2R-(2a,4aa,8aà)]	0.20
1-(Hydroxymethyl)-2,5,5,8A-tetramethyldecahydro-2-napthalenol	0.92
1-Hydroxy-4-oxo[10-2H,15-2H] borty-5(9)endiol	0.02
Unknown	0.68
Total	94.63

linalool (0.16%), 6-Methyl-5-hepten-2-one (0.05%), α-pinene oxide (0.02%) and limonene oxide (0.02%). In the sub aerial part of *C. flexuosus* the major compounds identified were citral (30.47%), eudesmol (17.82%), elemol (14.16%), dihydro isopropyl-8a-methyl azulene(11.08%), ë-cadinene (1.88%), junipene (1.36%), hydroxyalloaromadendrene, juniper camphor (1.12%) and à- elemene (1.04%).

Discussion

Essential oil content is a crucial criterion in determining the quality of *C.flexuosus* oil. The result indicated a higher percentage of essential oil from aerial parts compared to the essential oil obtained from the leaf part of *C.flexuosus* grown in India (Kulkarni, 1994; Padalia et al., 2011). The present study is the first report on the

essential oil composition from sub-aerial part of *C.flexuosus* containing citral as the major compound. The study shows that the essential oil of sub-aerial part not only serve as additional source of citral but contain other compounds like terpinene, elemene, junipene, cadinene, elemol, pulegone, eudesmol and juniper camphor used in perfumery, food, flavouring and in pharmaceutical industry.

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