STUDY ON ESSENTIAL OIL COMPOSITION OF THE ROOTS OF *CRASSOCEPHALUM CREPIDIODES* (BENTH.) S. MOORE

RAJESH K. JOSHI*

Department of Phytochemistry, Regional Medical Research Centre (Indian Council of Medical Research),
Belgaum, Karnataka-590 010, India

(Received: August 14, 2013 - Accepted: January 24, 2014)

ABSTRACT

The aim of the present study was to analyzed chemical constituents of the essential oil, obtained by hydro distillation of the roots of *Crassocephalum crepidioides* (Benth.) S. Moore. The oil was analyzed using gas chromatography equipped with flame ionization detector (GC-FID) and gas chromatography coupled with mass spectrometry (GC/MS). Forty-seven compounds were identified, representing 93.2 % of the total oil. The main constituents were (*E*)- β -farnesene (30.6%), α -humulene (10.3%), β -caryophyllene (7.2%), *cis*- β -guaiene (6.1%) and α -bulnesene (5.3%). The oil was constituted mainly of sesquiterpene hydrocarbons (77.6%), followed by oxygenated sesquiterpenes (8.7%), oxygenated monoterpenes (3.5%), monoterpene hydrocarbons (2.8%) and phenyl derivatives (0.6%). This is the first report on the essential oil composition of the roots of *C. crepidioides*.

Keywords: *Crassocephalum crepidioides* (Benth.) S. Moore, Essential oil composition, (*E*)- β -Farnesene, GC/MS.

INTRODUCTION

Crassocephalum crepidioides (Benth.) S. Moore (syn. *Gynura crepidioides* Benth.) commonly known as Thickhead or Fireweed is belongs to the family Asteraceae. The genus *Crassocephalum* is represented approximately by 100 species distributed in Asia, Africa, Australia, Malaysia, China, Nepal and Sri Lanka. In India, the genus *Crassocephalum* is represented by 8 species [1]. This plant is used as traditional medicine for treatment of cut wound and to cure diarrhea in various countries like India, Nepal and China [2-6]. It is a medicinal plant having free radical scavenging and hepatoprotective actions [7], antioxidant activity [8], and lipid peroxidation inhibitory effect [9]. There are few literature reports on the phytochemical analysis of this plant. Jacobine and jacoline very toxic pyrrolizidine alkaloids have been isolated from aerial parts of this species [10]. The essential oil of the flowers of the *C. crepidioides* has been reported from India to possess myrcene, β -phellandrene, dauca-5,8-diene, *trans*- β -farnesene and daucene as the major constituents, while the leaf oil was found to contain myrcene, β -phellandrene, *allo*-aromadendrene, α -muurolene and β -panasinsene as the major components [11]. Leaves oil of *C. crepidioides* from Cameroon were α -phellandrene, *p*-cymene, myrcene, limonene and (*E*)- β -ocimene as the major constituents [12]. In another report from Nigeria, the leaf oil contained α -caryophyllene, β -cubebene and α -farnesene, while thymol, α -caryophyllene and 4-cyclohexylbutyramide were found as the major constituents from stems oil of *C. crepidioides* [13]. To the best of author's knowledge, this is the first report on the chemical composition of the essential oil from the roots of *C. crepidioides*.

EXPERIMENTAL

Plant Material

The roots of *C. crepidioides* were collected in the month of September, 2011 from district Belgaum (Latitude N 15.88668; Longitude E 74.52353), Karnataka, India, at a height of 800 m. The plant herbarium and voucher specimen (RMRC-582) has been deposited in Regional Medical Research Centre (RMRC) Belgaum, Karnataka, India.

Isolation of Essential Oil

The fresh plant material (250 g) was subjected to hydro-distillation using Clevenger type apparatus for 3 h. The oil was collected and dried over anhydrous sodium sulfate and stored in sealed vials at -4°C until analysis. The oil yield was 0.03% (v/w).

GC-FID and GC-MS Analysis of the Essential Oil

The oil was analyzed by using a Varian 450 (TG-5, 30 m $\times$ 0.25 mm i.d., 0.25 μ m film thickness) Gas Chromatograph under the experimental conditions reported earlier [14,15]. The oven temperature was programmed from 60-220°C at 3°C/min, using nitrogen as carrier gas. The injector temperature was 230°C and the detector (FID) temperature 240°C. GC-MS utilized a Thermo Scientific Trace Ultra GC interfaced with a Thermo Scientific ITQ 1100 Mass Spectrometer fitted with a TG-5 (Thermo Scientific) fused silica capillary column (30 m $\times$ 0.25 mm; 0.25 μ m film thickness). The oven temperature range

was 60-220°C at 3°C /min using helium as carrier gas at 1.0 mL/min. The injector temperature was 230°C, and the injection volume 0.1 μ L in *n*-hexane, with a split ratio of 1:50. MS were taken at 70 eV with a mass range of *m/z* 40-450. All the experimental parameters were applied from those reported earlier [16, 17].

Identification of the Components

Identification of constituents were done on the basis of Retention Index (RI, determined with reference to homologous series of *n*-alkanes C₈-C₂₅, under identical experimental condition), MS library search (NIST 08 MS Library and WILEY MS 9th Edition), and by comparison with MS literature data [18]. The relative amounts of individual components were calculated based on GC peak area (FID response) without using correction factor.

RESULTS AND DISCUSSION

Forty-seven constituents were characterized and identified by GC-MS, comprising 93.2% of the total oil. The identified compounds are listed in Table 1 in elution order from the TG-5 column, along with the percentage composition of each component and its retention index. The major constituents of the oil were (*E*)- β -farnesene (30.6%) (Fig.1), α -humulene (10.3%), β -caryophyllene (7.2%), *cis*- β -guaiene (6.1%) and α -bulnesene (5.3%). The essential oil consists mainly of 18 sesquiterpene hydrocarbons (77.6%), followed by 12 oxygenated sesquiterpenes (8.7%), 1 oxygenated monoterpene (3.5%), 14 monoterpene hydrocarbons (2.8%) and 2 phenyl derivatives (0.6%). The composition of the essential oil often changes between different plant parts [19]. The difference in the complex composition of essential oils of one kind may sometimes be difficult to assign to specific chemotypes. The formation of essential oils depends on the tissue differentiation (secretory cells and excretion cavities, etc.) and on ontogenetic phase of the respective plant [20]. The secondary volatile metabolites of different parts of same plant virtually showed qualitative and quantitative differences of compounds reported in the essential oil composition from aerial parts and flowers of *Anaphalis contorta* [21-23], *Craniotome furcata* [24,25], *Chromolaena odorata* [26], *Coleus aromaticus* [27], *Tagetes minuta* [28], *Curcuma longa* [29], *Artemisia persica* [30], *Senecio belgaumensis* [31,32], *Prangos ferulacea* [33] and *Lantana camara* [34] and sometime derived different chemotypes from the roots of same plant [29,30,35]. The conclusion of this study revealed that the roots of *C. crepidioides* produced different chemotypes, namely, (*E*)- β -farnesene, α -humulene, β -caryophyllene, *cis*- β -guaiene and α -bulnesene, other than reported from the leaf, stem and flower parts oils (Table 2).

ACKNOWLEDGMENTS

The author is grateful to Indian Council of Medical Research, New Delhi, India for providing necessary facilities. Thanks also to Mr. Manjunath Patil, Lab Attendant for collection of plant material and extraction of oil.

Fig1. GC-TIC chromatogram of the essential oil of the roots of *C. crepidioides* and its major compound (*E*)- β -farnesene.

Table 1. Percentage composition of essential oil of the roots of *C. crepidioides*

Compound	RI	%	Identification*
Sabinene	979	t	RI, MS
β -Pinene	984	t	RI, MS
Myrcene	994	0.5	RI, MS
α -Phellandrene	1009	t	RI, MS
<i>p</i> -Cymene	1030	0.2	RI, MS
β -Phellandrene	1036	0.6	RI, MS
(<i>Z</i>)- β -Ocimene	1045	0.4	RI, MS
(<i>E</i>)- β -Ocimene	1056	0.2	RI, MS
γ -Terpinene	1064	t	RI, MS
Terpinolene	1093	t	RI, MS
Linalool	1105	0.1	RI, MS
Borneol	1170	0.3	RI, MS
Terpin-4-ol	1183	0.4	RI, MS
α -Terpineol	1195	0.1	RI, MS
Thymol, methyl ether	1242	t	RI, MS
Carvacrol, methyl ether	1251	0.6	RI, MS
Bornyl acetate	1293	3.5	RI, MS
Cyclosativene	1379	0.2	RI, MS
α -Copaene	1383	3.5	RI, MS
Isocomene	1392	0.2	RI, MS
β -Elemene	1397	3.4	RI, MS
Cyperene	1406	3.7	RI, MS
α -Gurjunene	1417	t	RI, MS
β -Caryophyllene	1428	7.2	RI, MS
2,5-Dimethoxy- <i>p</i> -cymene	1433	0.5	RI, MS
<i>trans</i> - α -Bergamotene	1441	0.5	RI, MS
α -Humulene	1460	10.3	RI, MS
(<i>E</i>)- β -Farnesene	1466	30.6	RI, MS
β -Chamigrene	1484	3.0	RI, MS
<i>cis</i> - β -Guaiene	1499	6.1	RI, MS
α -Selinene	1503	1.1	RI, MS
α -Murolene	1506	t	RI, MS
α -Bulnesene	1512	5.3	RI, MS
Cubebol	1525	0.8	RI, MS
δ -Cadinene	1533	1.7	RI, MS
Occidentalol	1557	0.2	RI, MS
Germacrene B	1572	0.3	RI, MS

Ledol	1577	t	RI, MS
Caryophyllene oxide	1590	1.2	RI, MS
Guaïol	1612	0.9	RI, MS
Hinesol	1651	2.8	RI, MS
Valerianol	1667	2.0	RI, MS
Occidentalol acetate	1691	t	RI, MS
Cedr-8-en-13-ol	1699	t	RI, MS
14-oxy- α -Murolene	1776	0.4	RI, MS
Cedrene-8,13-ol acetate	1809	0.2	RI, MS
Khusinol acetate	1831	0.2	RI, MS
Monoterpene hydrocarbons		2.8	
Oxygenated monoterpenes		3.5	
Sesquiterpene hydrocarbons		77.6	
Oxygenated sesquiterpenes		8.7	
Phenyl derivatives		0.6	
Total identified		93.2%	

RI=Retention index relative to C_8 - C_{25} *n*-alkanes on TG-5 column, MS=NIST and Wiley library and the literature, t=trace (<0.1%).

Table 2. Comparative analysis between major components of essential oil of leaf, stem flower and root of *C. crepidioides*.

Plant parts	Major compounds
Leaf	β -Phellandrene, <i>allo</i> -aromadendrene, α -murolene β -panasinsene [11], α -phellandrene, <i>p</i> -cymene, myrcene, limonene, (<i>E</i>)- β -ocimene [12], α -caryophyllene, β -cubebene and α -farnesene [13]
Stem	Thymol, α -caryophyllene and 4-cyclohexylbutyramide [13]
Flower	Myrcene, β -phellandrene, dauca-5,8-diene, <i>trans</i> - β -farnesene and daucene [11]
Root	(<i>E</i>)- β -Farnesene, α -humulene, β -caryophyllene, <i>cis</i> - β -guaïene and α -bulnesene

REFERENCES

- P. K. Hajra, R. R. Rao, D. K. Singh, B. P. Uniyal Flora of India. BSI Publication, Kolkata, India, 1995.
- S. R. Hynniewta, V. Kumar Indian J Tradit. Know. 7, 581, (2008).
- K. S. Rajbhandari Ethnobotany of Nepal. Ethnobotanical Society of Nepal C/o Central Department of Botany Kathmandu, Nepal, 2001.
- N. P. Manandhar Plants and People of Nepal. Timber Press, Portland, Oregon, 2002.
- S. R. Baral, P. P. Kurmi A Compendium of Medicinal Plants in Nepal. Mrs. Sharma R. 281 Maiju Bahal, Chabahill, Kathmandu-7, Nepal, 2006.
- E. Roeder, Pharmazie 55, 711, (2000).
- Y. Aniya, T. Koyama, C. Miyagi, M. Miyahira, C. Inomata, S. Kinoshita, T. Chiba, Biol. Pharm. Bull. 28, 19, (2005).
- O. A. Odukoya, S. A. Inya-Agha, F. A. Segun, M. O. Sofidiya, O. O. Ilori Am. J. Food Tech. 2, 169, (2007).
- S. O. Salawu, A. A. Akindahunsi, P. Cornuzzo, J. Pharmacol. Toxicol. 1, 429, (2006).
- Y. Asada, M. Shiraishi, T. Takeuchi, Y. Osawa, T. Furuya, Planta Med. 51, 539, (1985).
- R. K. Joshi, Int. J Nat. Prod. Res. 1, 19, (2011).
- P. H. A. Zollo, J. R. Kuïate, C. Menut, J. M. Bessiere, J. Essent Oil Res. 12, 533, (2000).
- I. A. Owokotomo, O. Ekundayo, I. A. Oladosu, S. A. Aboaba, Int. J. Chem. 4, 34, (2012).
- R. K. Joshi, Nat. Prod. Commun. 8, 401, (2013).
- R. K. Joshi, Natl. Acad. Sci. Lett. 36, 349, (2013).
- R. K. Joshi, Nat. Prod. Commun. 6, 1145, (2011).
- R. K. Joshi, J. Ethnopharmacol. 145, 621, (2013).

18. R. P. Adams, Identification of essential oil components by gas chromatography/mass spectrometry, 4th ed. Allured Publ. Corp., Carol Stream, IL, 2007.
19. C. B. Johnson, A. Kazantzis, M. Skoula, U. Mitteregger, J. Novak, *Phytochem. Anal.* 15, 286, (2004).
20. C. Franz, J. Novak (2010) CRC Press, New York, USA, 2010.
21. A. K. Sinha, M. S. Mehra, G. K. Sinha, *Indian Perfum.* 18, 35, (1974).
22. C. Pande, R. K. Joshi, S. S. Sammal, *J. Essent. Oil Res.* 20, 444, (2008).
23. R. K. Joshi, *Nat. Prod. Commun.* 8, 225, (2013).
24. R. K. Joshi, C. Pande, *Nat. Prod. Commun.* 3, 923, (2008).
25. R. K. Joshi, C. Pande, *J. Essent. Oil Res.* 21, 270, (2009).
26. R. K. Joshi, *J. Essent. Oil Bear. Pl.* 16, 71, (2013).
27. R. K. Joshi, V. Badakar, S. D. Kholkute, *Adv. Environ. Biol.* 5, 1307, (2011).
28. E. R. Chamorro, G. Ballerini, A. F. Sequeira, G. A. Velasco, M. F. Zalazar, *J. Argent. Chem. Soc.* 96, 80, (2008).
29. N. K. Leela, A. Tava, P. M. Shahj, S. P. John, B. Chempakam, *Add Pharm.* 52, 137, (2002).
30. B. F. Mirjalili, M. H. H. Meybody, M. M. Ardakani, A. Rustaiyan, N. Ameri, S. Masoudi, A. Bamoniri, *J. Essent. Oil Res.* 18, 544, (2006).
31. R. K. Joshi, *Nat. Prod. Commun.* 6, 1145, (2011).
32. R. K. Joshi, *Chem. Nat. Compd.* 47, 1010, (2012).
33. M. T. Akbari, A. Esmaili, A. H. Zarea, N. Saad, F. Bagheri, *Bulg. Chem. Commun.* 42, 36, (2010).
34. R. K. Joshi, *J. Biol. Act. Prod. Natr.* 2, 135, (2012).
35. R. K. Joshi, *J. Chem.* 2013, 1, (2013).