

Original Article

Volatile constituents of the peel and leaf of *Citrus aurantium* L. cultivated in the north of Iran

Open Access

Boshra Azadi¹, Bahman Nickavar², Gholamreza Amin^{1*}

1. Department of Pharmacognosy, Pharmaceutical Sciences Branch, Islamic Azad University, Tehran, Iran

2. Department of Pharmacognosy, Faculty of Pharmacy, Shahid Beheshti University of Medical Sciences, Tehran, Iran

Abstract

The essential oil constituents of the peel and leaf of *Citrus aurantium* L. (Rutaceae) grown in the north of Iran, were analyzed by GC and GC/MS. Fourteen components representing 99.6% of the leaf oil were identified. The major compounds were linalool (39.4%), linalyl acetate (38.8%) and α -terpineol (7.2%). Twenty constituents consisting 99.4% of the peel oil were identified. The main components were limonene (91.3%), β -myrcene (3.0%) and linalool (1.1%).

Key words: *Citrus aurantium* L., Essential oil, Rutaceae, Limonene, Linalool, Linalyl acetate

*Correspondence to: Gholamreza Amin, Ph.D, Professor of Pharmacognosy, Pharmacognosy Department, Pharmaceutical Sciences Branch, Islamic Azad University, Tehran, Iran.

Tel.: +98 21 22640051 Fax: +98 21 22602059

Email address: gh_amin@yahoo.com

1. Introduction

The genus Citrus (Rutaceae) is found in the temperate and semitropical areas of Iran and many species of this genus are cultivated in this area (Ghahreman, 1993)(Salehi, Mohammadi et al. 2008). Citrus aurantium L. or sour orange is a tree up to 6 m. height with leathery leaves, white and aromatic flowers, globular and orange coloured fruit which first originated in the north Indian areas (Mozaffarian, 2003; Evans, 2006)(Schiff 1980; Mahmoudi, Seyedabadi et al. 2011). It is locally named Narenjand its flowers (BaharNarenj) used as a sedative agent in the folk medicine of Iran (Amin, 2005)(Fazeli, Amin et al. 2007). Because of wide uses of Citrus species, the cultivation of those was extended in whole temperate areas of Iran (Zargari, 1992) (Sahraei, Shams et al. 2007). According to the recent studies on Citrus aurantium L., the peel oil has antimicrobial (Sonbol et al., 1992), antifungal (Ramadan et al., 1996)(Martin and Ernst 2004), insecticide (Mwaiko, 1992) (Kamaraj, Rahuman et al. 2008), antioxidative (Song et al., 2001)(Shahidi and Zhong 2005) and cardiovascular (Occhiuto and

Circosta,1996)(Occhiuto and Circosta 1996) effects.

The literature survey revealed that linalool, linalyl acetate and α -terpineol were the major compounds in the leaf oil (Baaliouamen and Meklati, 1986; Calvarano,1968; Di Giacomo and Romeo,1974; Karawya et al.,1970) (Karawya, Hashim et al. 1974; Guenther, Gilbertson et al. 1977; Fishman, Erdmann et al. 1981; Gogorcena and Ortiz 1989) and the main components of the peel oil were limonene and myrcene, in many countries (Boelens and Jimenez, 1989; Dugo and Giacomo, 2002; Lota et al.,2001; Samahy et al.,1982)(Dugo, Mondello et al. 1997; Lin and Rouseff 2001; Lota, de Rocca Serra et al. 2001; Pérez-López, Saura et al. 2006) but there was no report on volatile constituents of Citrus aurantium L. peel and leaf cultivated in the north of Iran.

2. Materials and Methods

Plant material

Citrus aurantium L. leaves and fruits were collected from Sari in the north of Iran, in April and November 2002, respectively. Samples were authenticated by Prof. Golamreza Amin

Table 1: Chemical composition of Citrus aurantium L. leaf oil

No.	Compound	RI ^a	Percentage
1	α -Pinene	935	0.3
2	Sabinene	972	0.5
3	β -Pinene	976	3.8
4	Myrcene	988	0.7
5	Limonene	1024	0.3
6	cis-Linalool oxide	1064	0.2
7	Linalool L	1095	39.4
8	α -Terpineol	1173	7.2
9	trans-Geraniol	1203	0.8
10	Linalyl acetate	1228	38.8
11	Geranial	1240	0.4
12	Neryl acetate	1315	2.5
13	Geranyl acetate	1332	4.5
14	trans-Caryophyllene	1368	0.2

^a RI: retention indices on DB-5 capillary column

and a voucher specimen (No. 1041-HPAU) has been deposited at the herbarium of the Pharmacognosy Department, Pharmaceutical Sciences Branch, Islamic Azad University, Tehran, Iran.

Oil isolation

The fresh crushed leaves and peels of *Citrus aurantium* L. were separately subjected to hydrodistillation using a Clevenger-type apparatus for 4 hrs. The obtained essential oils were dried over anhydrous sodium sulphate and stored at 4-6°C.

GC and GC/MS analysis

The leaf essential oil was analyzed by GC and GC/MS using a Hewlett-Packard 6890 gas

chromatograph with DB-5 capillary column (30 m x 0.25 mm; film thickness 0.25 mm). The carrier gas was helium with a flow rate of 1 ml/min. The column temperature was programmed from 60°C to 220°C at 60°C/min. The gas chromatograph was coupled to a Hewlett-Packard 5973 mass selective detector. The MS was operated at 70 eV ionization energy.

The retention indices were calculated by using retention times of n-alkanes that were injected after the essential oil at the same conditions. The components were identified by comparison of retention indices with those reported in the literatures and also by comparison of their mass spectra with the published mass spectra or Wiley library (Adams, 2001; Massada, 1976)

Table 2: Chemical composition of *Citrus aurantium* L. peel oil

No.	Compound	RI ^a	Percentage
1	Hexanal	773	0.1
2	α-Pinene	923	1.0
3	β-Pinene	960	0.9
4	β-Myrcene	978	3.0
5	Limonene	1024	91.3
6	(E)-β-Ocimene	1053	0.5
7	Octanol	1062	0.2
8	trans-Linalool oxide	1072	trace ^b
9	Nonanal	1082	trace
10	Linalool	1090	1.1
11	4-Terpineol	1152	trace
12	α-Terpineol	1162	0.2
13	Decanal	1181	0.2
14	Nerol	1204	0.1
15	(z)-Citral	1207	0.1
16	trans-Geraniol	1232	0.1
17	Linalyl acetate	1238	0.5
18	Neryl acetate	1336	trace
19	Geranyl acetate	1355	0.1
20	Nerolidol	1536	trace

^aRI: retention indices on DB-1 capillary column

^btrace: The values under 0.05% were considered as a trace.

(Baaliouamer, Meklati et al. 1985; Tirillini, Pagiotti et al. 2009). Relative percentage amounts were calculated from peaks total area by apparatus software.

GC and GC/MS analysis of the peel oil was performed on a Thermoquest 2000 system with DB-1 capillary column (30 m x 0.25mm; film thickness 0.1mm). The carrier gas was helium with a flow rate of 1.5 ml/min. The column temperature was programmed from 50°C to 265°C at 2.5°C/min. The MS was taken at 70eV. Identifying the compounds was carried out as same as the leaf method.

3. Results and Discussion

The fresh leaves of *Citrus aurantium* L. yielded 0.19% V/W of a clear yellow volatile oil with a fresh sweet and neroli odor.

Fourteen compounds representing 99.6% of the total oil were identified. The detected constituents and their percentage are shown in Table 1.

The major components were linalool (39.4%), linalyl acetate (38.8%) and α -terpineol (7.2%). The leaf essential oil contained 47.6% alcohols and 45.8% esters.

The fresh peels of *Citrus aurantium* L. yielded 1.95% V/W of a pale yellow volatile oil with a strong pleasant odor.

Analyzing of the peel oil showed twenty compounds which are given in Table 2.

The identified components were represented 99.4% of the total oil. *Citrus aurantium* L. peel oil contained 93.2% cyclic monoterpenes with limonene (91.3%) as the principle constituent. Another compound which presented in appreciable amount was β -myrcene (3.0%).

This research on peel and leaf essential oils of *Citrus aurantium* L. confirms the previous reports on this species from the other countries. According to the references, the peel oil quality is attributed to limonene content and the presence of 90% limonene is the optimum value (BPC, 1973). This investigation shows that *Citrus aurantium* L. peel oil cultivated in

the north of Iran with 91.3% limonene has a high quality for industrial purpose.

Conflict of interests : None declared.

4. References

Adams RP. Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectroscopy. Illinois, USA, Allured Publishing Corporation, 2001.

Amin Gh. Popular Medicinal Plants of Iran. Tehran, Tehran University of Medical Sciences Publication, 2005: 82 & 142.

Baaliouamen A, Meklati BY. Analysis of bitter orange petitgrain essential oil by combined gas chromatography-mass spectrometry. *Agricbiolchem* 1986;50(8): 2111-2114.

Boelens MH, Jimenez R. The chemical composition of the peel oil from unripe and ripe fruits of bitter orange, *Citrus aurantium* L. *Flavour Fragrance J* 1989; 4(3): 139-142.

Calvarano I. Italian petitgrain oil. Bigarade and bergamot petitgrain oils. *Essenze Driv Agrum* 1968; 38(1-2): 31-48.

Dugo G, Giacomo A. *Citrus*. 1st ed. London, Tyler & Francis, 2002.

Di Giacomo A, Romeo G. Analysis of Italian petitgrains by IR spectroscopy. *Essenze Drive Agrum* 1974; 44(3): 217-235.

Evans WC. Trease and Evans Pharmacognosy. 15th ed. London, WB Saunders Company, 2006: 266-267.

Ghahreman A. *Plant Systematic*. Tehran, Tehran University Press, 1993: Vol. 2.

Karawya MS, Balbaa SI, Hifnawy MS. Leaf essential oils of bitter orange and bergamot growing in Egypt. *AmerPerfumCosmet*

1970;85(11):29-32.

Lota M, De Rocca Serra D, Jacquemond C, Tomi F, Casanova J. Chemical variability of peel and leaf essential oil *Citrus aurantium*. *Flavour Fragrance J* 2001; 16(2): 89-96.

Massada Y. *Analysis of Essential Oil by Gas Chromatography and Mass Spectrometry*. New York, John Wiley & Sons Inc., 1976.

Mozaffarian V. *A Dictionary of Iranian Plant Names*. Tehran, Farhang Moaser, 2003:131.

Mwaiko GL. Citrus peel oil extracts as mosquito larvae insecticides. *East Afr Med J* 1992;69(4): 223-226.

Occhiuto F, Circosta C. Cardiovascular properties of the non-volatile total residue from the essential oil of *Citrus aurantium*. *Int J Pharmacogn* 1996;34(2): 128-133.

Pharmaceutical Society of Great Britain. *British Pharmaceutical Codex*. London, The Pharmaceutical Press, 1973:558.

Ramadan W, Mourad B, Ibrahim S, Sonbol F. Oil of bitter orange: new topical antifungal agent. *Int J Dermatol* 1996; 35(6): 448-449.

Samahy SK, Askar A, Fadeel MG. Quantitative analysis of some Citrus peel oils. *RiechstAromenKosmet* 1982; 32(3): 68-70.

Sonbol F, Ibrahim SM, Mohamed BM. Antimicrobial activity of oil of bitter orange. *Alexandria J Pharm Sci* 1992;9(2): 107-109.

Song HS, Ukeda H, Sawamura M. Antioxidative activities of Citrus peel essential oils and their components against linoleic acid oxidation. *FoodSciTechnol Res* 2001; 7(1): 50-56.

Zargari A. *Medicinal Plants*. 6th ed. Tehran, Tehran University Press, 1992: Vol.1.