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RESEARCH REPORT

Italian *Citrus* Petitgrain Oils. Part II. Composition of Mandarin Petitgrain Oil[1]

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Abstract

The chemical composition of Italian industrial mandarin petitgrain oil was examined using GC on two capillary columns and a fully automated "on-line" HPLC-GC/MS (ITD) system and a GC/MS (quadrupole) system. Methyl N-methyl anthranilate was found to be the main component representing about 50% of the whole oil. γ-Terpinene (23.9-28.5%) was the main monoterpene hydrocarbon, followed by limonene (7.2-12.6%) and p-cymene (3.0-5.2%). Alcohols were found to represent only about 1% of the oil, while aldehydes were present only as traces.

Key Word Index

Citrus reticulata, Rutaceae, mandarin leaf oil, mandarin petitgrain oil, essential oil composition, methyl N-methyl anthranilate, γ-terpinene, limonene.

Introduction

In a previous paper the composition of bitter orange petitgrain oil (1) was reported. The analyses were carried out by a fully automated HPLC-GC/MS (ITD) system which allowed the LC separation between hydrocarbon and oxygenated compounds and the following analysis of the two fractions, by GC/MS (quadrupole) and by GC on SE-52 and Carbowax 20 M columns. Some commercial MS-libraries and a home-made library equipped with Linear Retention Indices were used interactively with MS data for peak identification (2). In this paper, the results of the composition of Italian industrial mandarin petitgrain oils, obtained using the same techniques used for bitter orange petitgrain oil, are reported.

Mandarin petitgrain oil world production is rather limited and it is almost exclusively carried out in Italy (3). Lawrence (3) has reviewed most of the papers concerning mandarin petitgrain oil (4-23). In 1974, Di Giacomo (13) reported an up-to-date list of the components identified in mandarin petitgrain oil. Almost all the studies were performed on laboratory produced samples where the oil was isolated by steam distillation (7,8,14,17-20,22,23) or by solvent extraction (10-12,21). Some of these reports deal with a comparison of different varieties of mandarin (7,16,17,20-22), while others correlate the composition of the oil with the period of harvesting (8,11,18). The composition of Italian mandarin petitgrain oil is reported only in a paper of Calvarano (9), where the different classes of compounds were separately analyzed. In this report, the total amount of each class was not reported, so that it was not possible to calculate the percentage content of each component, except for monoterpene hydrocarbons, methyl anthranilate and methyl N-methyl anthranilate.

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Table I shows most of the results on the composition of mandarin petitgrain oil reported in the literature. Moreover, some pertinent information such as the botanical origin or cultivar origin noted by the authors is included. Table I reveals that considerable qualitative and quantitative differences among data reported can be found in the literature. Fleisher and Fleisher (20) identified many sesquiterpene hydrocarbons, alcohols and esters not normally found in mandarin petitgrain oils in some

Table I. Quantitative composition (percentage) of mandarin petitgrain oil reported in the literature

Monoterpene hydrocarbons					
Camphene	0.02 (9)	0.2 (10)	0-0.3 (11)	0.1 (12)	0-tr (20)
	0.6-48.0 (21)	0.37 (23)			
δ-3-Carene	0.11 (9)	tr (12)	47.36 (14)	0-10 (16)	0.04 (23)
p-Cymene	3.43 (9)	22.3 (10)	17.7-22.7 (11)	0.6 (12)	0-4 (16)
	14.0-41.1 (18)	16.29 (19)	tr-6.54 (20)	0-9.1 (21)	2.35 (23)
α-Fenchene	0-1 (16)				
Limonene	35 (6)	0.8-1.9 (8)	8.22 (9)	2.9(10)	3.0-5.6 (11)
	1.2 (12)	3.35 (14)	1-6 (16)	1.99-4.67 (17)	2.8-4.8 (18)
	3.63 (19)	0.23-4.91 (20)	0-3.2 (21)	8.32 (23)	
Myrcene	0.3-0.6 (8)	0.88 (9)	0.3 (10)	0.2-0.8 (11)	2.2 (12)
	3.31 (14)	0-2 (16)	1.93-3.17 (17)	0.3-0.7 (18)	0.63 (19)
	0-2.00 (20)	0-2.7 (22)	0.05 (23)		
Ocimene	2.8-8.2 (8)[a]	0.68 (9)[a]	5.75 (14)[a]	0-5 (16)[a]	0-19.8 (22)[a]
(E)-β-Ocimene	2.1 (12)	1.74 (19)	7.87 (23)		
(Z)-β-Ocimene	0-4.16 (20)				
α-Phellandrene	0.12 (9)	0-12 (16)	0.49 (19)	0-0.05 (20)	0-2.4 (21)
β-Phellandrene	0.2-0.5 (8)	1.4 (12)	0-1.94 (22)		
α-Pinene	1.1-2.7 (8)[b]	2.16 (9)	3.5 (10)[b]	3.6-5.9 (11)[b]	1.3 (12)
	1.52 (14)	3-5 (16)	0.69-1.40 (17)	1.3-2.2 (18)	3.91 (19)
	0-1.83 (20)	2.66 (23)			
β-Pinene	1.4-2.6 (8)	1.86 (9)	9.1 (10)[c]	9.3-13.2 (11)[c]	1.4 (12)
	0.02 (14)	tr-2 (16)	5.8-20.2 (18)	4.71 (19)	0.02-1.51(20)
	0-24 (22)	0.86 (23)			
Sabinene	tr (9)	9.1 (10)[c]	9.3-13.2 (11)[c]	23.6 (12)	1.85 (14)
	0-20 (16)	0.4-0.9 (18)	0.57 (19)	0-27.20 (20)	4.1-16.5 (22)
	2.81 (23)				
α-Terpinene	0.1-0.3 (8)	0.17 (9)	0.1 (10)	0.1 (11)	0.1 (12)
	0.1-0.3 (18)	0.07-0.72 (20)	1.95 (23)		
γ-Terpinene	4.3-10.0 (8)	13.74 (9)	21.0 (10)	19.5-21.5 (11)	0.8 (12)
	11.8-38.3 (18)	20.15 (19)	0-12.60 (20)	0-21.8 (21)	0-6.6 (22)
	1.20 (23)				
Terpinolene	0.7-1.3 (8)	0.59 (9)	0.1 (12)	1.62 (19)	0-0.53 (20)
	1.49 (23)				
α-Thujene	1.1-2.7 (8)[b]	0.01 (9)	3.5 (10)[b]	3.6-5.9 (11)[b]	0.4 (12)
	0-0.78 (20)	1.26 (23)			
Sesquiterpene hydrocarbons					
α-Bergamotene	0.11 (23)				
trans-β-Bergamotene	0-0.03 (19)				
Bicyclogermacrene	0-0.54 (19)				
β-Bisabolene	0.4-4.0 (21)				
δ-Cadinene	0.08 (23)				
γ-Cadinene	0-0.47 (19)				
β-Caryophyllene	0.6 (10)	0.6-2.1 (11)	0.2 (12)	5.08 (14)	0-tr (16)
	0.6-10.2 (18)	0.09-5.44 (20)	1.4-8.4 (21)	0.24 (23)	

Table I. Continued

α-Copaene	0-0.17 (20)				
α-Elemene	2.5-4.0 (21)				
β-Elemene	1.3 (10)[d]	1.3-5.0 (11)[d]	tr (12)	11.0-25.4 (18)	0-1.28 (20)
	2.4-4.4 (21)				
δ-Elemene	0.22 (23)				
α-Farnesene	0-1.00 (20)				
(E)-β-Farnesene	0.2 (12)	0.72 (14)	0-tr (16)	0-0.82 (20)	
Germacrene B	0.10 (23)				
Germacrene D	0.17 (23)				
Humulene	6.1 (10)[a,e]	0.5-6.2 (11)[a,e]	0.1 (12)	0-tr (16)	
α-Humulene	tr-0.55 (20)	0.08 (23)			
Longifolene	0.23 (23)				
α-Muurolene	0-0.32 (20)	0.08 (23)			
β-Selinene	1.4 (10)	0.4-4.7 (11)	tr (12)	0.2-1.3 (18)	
Valencene	0-0.05 (20)				
Viridiflorene	0-0.31 (20)				
Alcohols					
α-Bisabolol	0.29 (23)				
δ-Cadinol	0.06 (23)				
cis-Carveol	0-0.06 (20)				
trans-Carveol	0-0.04 (20)				
Citronellol	1.25 (14)	0.70-7.78 (17)	0.1-0.5 (18)	5.19 (19)	0-2.60 (20)
p-Cymen-8-ol	tr-0.10 (20)				
1,2-Dihydrolinalool	0.03 (23)				
Geraniol	0.1-0.2 (8)	0.5 (10)	0.2-0.5 (11)	0.1 (12)	0.1-0.4 (18)
	0.35 (19)	0-0.05 (20)	0.16 (23)		
(E)-2-Hexenol	0.2 (10)	0-0.5 (11)	tr (12)		
(Z)-3-Hexenol	1.5 (10)	1.3-5.3 (11)	tr (12)	0-0.33 (20)	
Isopulegol	tr (12)	0-2 (16)			
Linalool	2 (6)	52-78 (8)	21.6 (10)	19.7-23.7 (11)	59.2 (12)
	21.82 (14)	0-35 (16)	16.69-27.13 (17)	3.4-15.2 (18)	9.55 (19)
	0.11-55.10 (20)	2.1-6.4 (21)	6.1-35.9 (22)	52.66 (23)	
Myrtenol	0-0.12 (20)				
Nerol	0.1 (12)	tr (14)	0.21 (19)		
Nerolidol	0-0.04 (20)				
Nonanol	0.19 (23)				
(Z)-2-Pentenol	0-tr (20)				
β-Phenylethanol	0-0.03 (20)				
cis-Sabinene hydrate	0-0.21 (20)	0.08 (23)			
trans-Sabinene hydrate	0-2.79 (20)				
Sabinol	0-tr (20)				
Terpinen-1-ol	0-0.02 (20)				
Terpinen-4-ol	1.3 (10)[d]	1.3-5.0 (11)[d]	0.5 (12)	0-25 (16)	7.13 (19)
	0-6.11 (20)				
α-Terpineol	1 (6)	0.2-0.6 (8)[f]	6.1 (10)[e]	0.5-6.2 (11)[e]	1.2 (12)
	0-3 (16)	0.4-5.6 (18)	0.85 (19)	0.24-1.30 (20)	0.06 (23)
Thymol	2.6-6.9 (8)	0.3 (12)	0-14.43 (20)	0-4.9 (22)	
Aldehydes					
Benzaldehyde	0-0.02 (20)				
Citronellal	0.1 (12)	0.07 (14)	0.87-6.67 (17)	0-0.25 (20)	
Decanal	tr (12)	0-0.03 (20)			
Dodecanal	tr (12)				
Geranial	0.2 (10)	0.2-0.5 (11)	0.1 (12)	0.07 (14)	0.83 (19)
	7.45 (23)				

Table I. Continued

(E)-2-Hexanal	3.6 (10)	0.7-3.7 (11)	tr (12)		
Neral	0.2-0.6 (8)[f] 6.05 (23)	tr (12)	0.78 (14)	0.3-0.6 (18)	0.36 (19)
Nonanal	tr (12)				
α-Sinensal	0-0.93 (20)	0-1.1 (22)[g]	0.50 (23)		
β-Sinensal	0-2.98 (20)	0-1.1 (22)[g]			
Esters					
Bornyl acetate	0.24 (19)				
Citronellyl formate	0-2.56 (20)	0.43 (23)			
Ethyl-2-butenoate	0-0.04 (20)				
Ethyl decanoate	0-0.14 (20)				
Ethyl heptadecanoate	tr-0.31 (20)				
Ethyl laurate	0-0.06 (20)				
Ethyl linoleate	0.39-1.78 (20)				
Ethyl linolenate	0.68-6.88 (20)				
Ethyl myristate	0.05-0.88 (20)				
Ethyl nonanoate	0-0.01 (20)				
Ethyl octanoate	0-0.04 (20)				
Ethyl palmitate	1.83-7.22 (20)				
Ethyl pentadecanoate	0-0.06 (20)				
Ethyl phenyl acetate	0-0.06 (20)				
Ethyl stearate	tr-0.87 (20)				
(Z)-3-Hexenyl formate	0-0.11 (20)				
Geranyl acetate	0.1 (12)	2.13 (19)			
Linalyl acetate	5 (6)	0-tr (16)			
Methyl anthranilate	0-3 (16)				
Methyl N-methyl anthranilate	10-15 (6)	0-60 (16)	0-65.71 (20)		
Methyl salicylate	0-1 (20)				
Octenyl acetate	0.36 (19)				
Oxides					
Caryophyllene oxide	0.42 (19)	0-2.10 (20)			
Humulene oxide	0-0.07 (20)				
cis-Limonene oxide	0.59 (23)				
cis-Linalool oxide	0-0.49 (20)	0-39.9 (22)[h]			
trans-Linalool oxide	0-0.29 (17)	0-39.9 (22)[h]			
Others					
β-Amyrin	4.9-14.2 (21)				
Anethole	0-0.03 (20)				
1,4-Cineole	0.27 (23)				
1,8-Cineole	3.82 (12)	0.70 (23)			
α-p-Dimethyl styrene	1.8 (10)	1.8-3.6 (11)	tr (12)		
Methyl thymol	1.1-16 (8)	0.3 (12)	0-4.63 (20)		
Phytol	0.09-1.29 (20)	8.8-18.3 (21)	0.13 (23)		
p-isoPropenyl toluene	0.6-1.1 (8)				

[a]correct isomer not characterized; [b]α-Pinene + α-Thujene; [c]β-Pinene + Sabinene; [d]β-Elemene + Terpinen-4-ol; [e]Humulene + α-Terpineol; [f]Neral + α-Terpineol; [g]α-Sinensal + β-Sinensal; [h]cis- + trans-Linalool oxide

Appendix to Table I: Details on the references on the composition of mandarin leaf oil.
Industry steam-distilled oils (4-6,9,15). Laboratory steam-distilled oils (7,8,14,17-20,22,23). Laboratory solvent extracted oils (10-12,21). Qualitative data (4,5,7,13,15). Quantitative data (6,8,12,14,16-22). Biogenesis/Ontogenesis (7,8,11,18). Chemotaxonomy (10-12,16,17,20-22). *Citrus reticulata* Blanco (7-9,12,14,16,19,20,22,23), cv. Dancy (7,8); cv. Murcott (21); cv. Kimow (14); cv. Balady, Clemantin, Suntara (16); cv. Satsuma (19); cv. Balady, Yussuf Effendi, Dancy, Maya, Clementine, Michal, Nectarine (20). *Citrus unshiu* Marcovitch (10,11,18,21,22). cv. Hayashi (21); cv. Miacawa, Okissu (22). *Citrus clementina* Hort. ex Tanaka, cv. Arrufatina, Clemenules, Clemenville, Esbal, Fina, Guillermina, Hernandina, Monreal, Oroval, Tomatera (17). *Citrus tangerine* Hort. ex Tanaka, cv. Dahongpao (22).

mandarin cultivars found in Israel. In addition, these same authors found that some compounds such as linalool (0.11-55.10%), methyl N-methyl anthranilate (0-65.71%) and sabinene (0-27.20%) varied considerably with different cultivars. Other compounds for which wide ranges have been reported were β-pinene (0 to 24%) (22) and δ-3-carene (0-47.36%) (14). Kekelidze et al. (18) report a value of 25% for β-elemene, a compound which is often either absent or present only as a trace component. Attaway (8) reported that the linalool content ranged from 52% to 78%, while it has been considered previously as being absent (16) or only present in a small amount (6,18,20).

Fleisher and Fleisher (20) and Lin and Hua (22) reported α- and β-sinensal as components of mandarin petitgrain oil. These two compounds are characteristic components of sweet orange petitgrain oil (24) and have not been previously identified in mandarin petitgrain oil. On the whole, data reported in the literature are not sufficient to characterize mandarin petitgrain oil.

Experimental

The analyses were performed on five samples of mandarin petitgrain oil. All the samples were isolated under our supervision, in a factory of a Sicilian oil producer in the spring of 1994. Each sample was obtained from 450 kg of leaves. The yield was about 2‰.

As for bitter orange petitgrain oil analysis (1), the LC-GC/MS (ITD) system was equipped with a SE-52 capillary column; for the analysis with the GC/MS (quadrupole) and GC systems, either SE-52 or Carbowax 20 M capillary columns were used as described previously (1).

Results and Discussion

Figure 1 reports the total ion current chromatograms of a mandarin petitgrain oil and its correspondent fractions obtained by LC-pre-separation. Figures 2 and 3 report the GC/MS (quadrupole) chromatograms using a Carbowax 20 M and a SE-52 column, respectively. Table II reports peaks identified by LC-GC/MS (ITD), by GC/MS (quadrupole) on Carbowax column and by GC/MS (quadrupole) on SE-52 column. Table III contains the quantitative composition of the analyzed mandarin petitgrain oil samples. These data refer to the results obtained by GC with SE-52 and Carbowax columns, and to those of the identification carried out by GC/MS (quadrupole) and LC-GC/MS (ITD).

The GC/MS (ITD) analysis of the fractions obtained by LC-pre-separation allowed a more certain identification of compounds present in low amounts, such as tricyclene, 6-methyl-5-hepten-2-one, 1,8-cineole, cis-sabinene hydrate, cis-linalool oxide (furanoid form), octanol, cis-limonene oxide, p-cymen-8-ol, methyl anthranilate, α-terpinyl acetate, (E,E)-α-farnesene and δ-cadinene. Analyses carried out with the Carbowax 20 M column, allowed the resolution of the following couples of compounds: limonene/β-phellandrene, terpinolene/p-cymenene and α-selinene/bicyclogermacrene, which were not resolved on the SE-52 column.

As can be seen from Table II and Figures 1-3, 66 components were identified, representing more than 99.5% of the whole oil. Tricyclene, 6-methyl-5-hepten-2-one, octanal, o-cymene, octanol, p-cymenene, p-mentha-1,3,8-triene, cis- and trans-p-menth-2-en-1-ol, carvacrol, α-terpinyl acetate, neryl acetate, methyl N-dimethyl anthranilate, α-selinene, (Z)-3-hexenylbenzoate and spathulenol were identified for the first time in mandarin petitgrain oil. Many components, previously reported as present (Table I), were not found.

Monoterpene hydrocarbons represented from 45% to 54% of the whole oil, while sesquiterpene hydrocarbons ranged from 1.0% to 1.7%. The most abundant monoterpene hydrocarbon was γ-terpinene (24-28%), followed by limonene (7-13%). β-Caryophyllene, which represented about 70% of the whole sesquiterpene fraction, was the main sesquiterpene hydrocarbon. Esters were the major class of substances which contributed to the total oxygenated constituents content. In particular, methyl N-methyl anthranilate, which represented from 42% to 52% of the whole oil, was the main component. Alcohols were present in quantities varying from 0.8% to 2.0%, while aldehydes were present only in small amounts (0.04-0.22%).

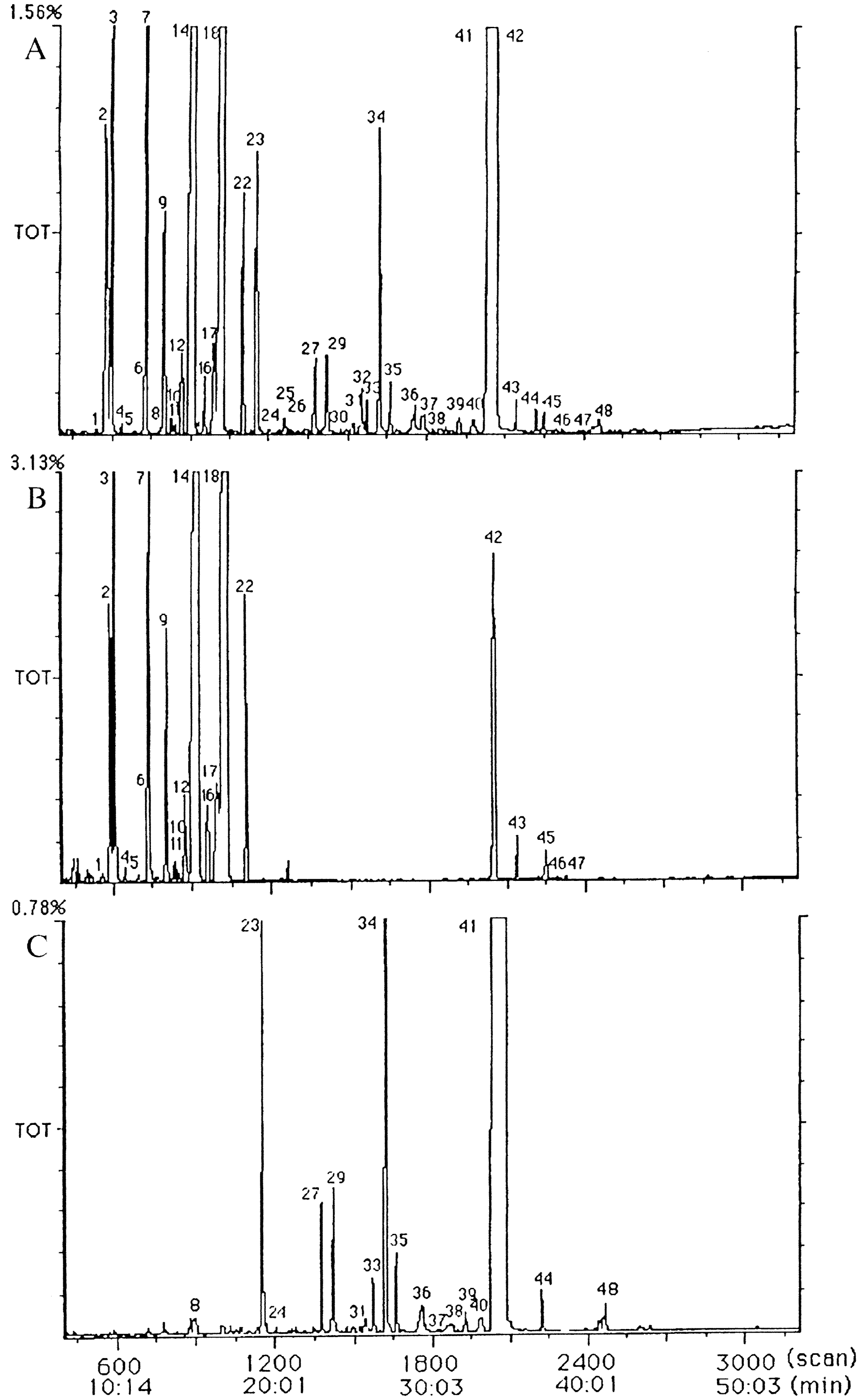

Figure 1. Total ion current chromatograms of a mandarin petitgrain oil and fractions from its LC separation. GC column, SE-52, 30 m. For peak identification, see Table II

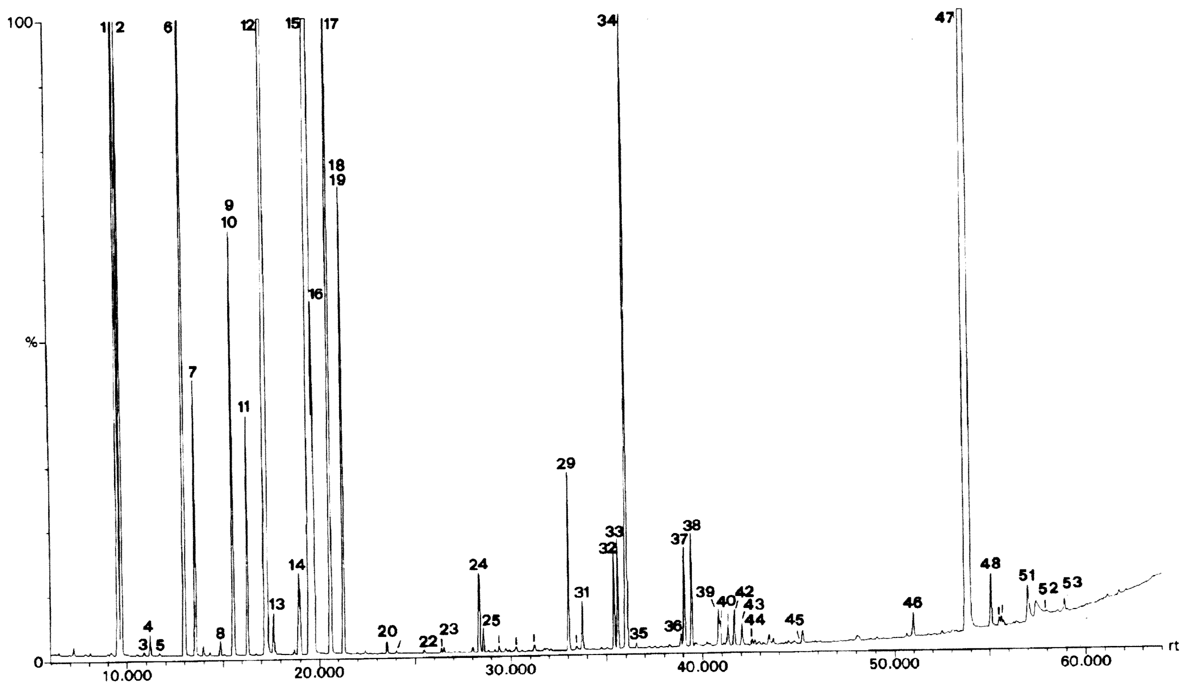

Figure 2. Total ion current chromatograms of a mandarin petitgrain oil. GC column, Carbowax 20M, 60 m. For peak identification, see Table II

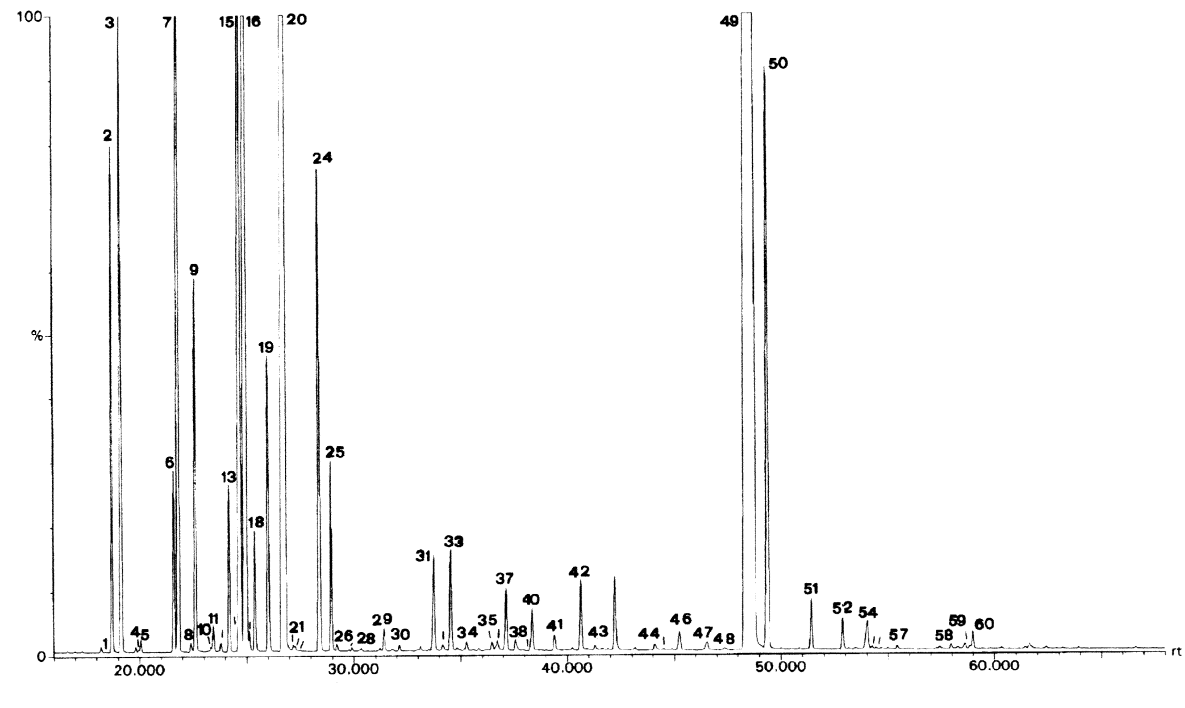

Figure 3. Total ion current chromatograms of a mandarin petitgrain oil. GC column, SE-52, 60 m. For peak identification, see Table II

Table II. Compound identification by LC-GC/MS (ITD), by GC/MS (quadrupole) on Carbowax column and by GC/MS (quadrupole) on SE-52 column

Compounds	LC-GC/MS Peak in Figure 1	GC/MS on Carbowax column Peak in Figure 2	GC/MS on SE-52 column Peak in Figure 3
hexanal		5	
(Z)-3-hexenol		22	
hexanol		21	
tricyclene	1		1
α-thujene	2	2	2
α-pinene	3	1	3
α-fenchene	4	3	4
camphene	5	4	5
sabinene	6	7	6
β-pinene	7	6	7
6-methyl-5-hepten-2-one	8	20	8
myrcene	9	9	9
octanal		19	10
α-phellandrene	10	10	11
δ-3-carene	11	8	12
α-terpinene	12	11	13
o-cymene			14
p-cymene	13	17	15
limonene	14	12	16
β-phellandrene		13	
1,8-cineole	15		17
(Z)-β-ocimene	16	14	18
(E)-β-ocimene	17	16	19
γ-terpinene	18	15	20
cis-sabinene hydrate	19	26	21
octanol	21	30	22
cis-linalool oxide (furanoid)	20		23
terpinolene	22	18	24
p-cymenene		24	
linalool	23	29	25
nonanal	24	23	26
p-mentha-1,3,8-triene		25	27
cis-p-menth-2-en-1-ol		35	28
cis-limonene oxide	25		
trans-p-menth-2-en-1-ol	26		29
citronellal		27	30
terpinen-4-ol	27	33	31
p-cymen-8-ol	28		32
α-terpineol	29	38	33
decanal	30	28	34
citronellol	31		35
nerol	32		36
methyl thymol		32	37
neral	33	36	38
geraniol		45	39

Table II. Continued

Compounds	LC-GC/MS Peak in Figure 1	GC/MS on Carbowax column Peak in Figure 2	GC/MS on SE-52 column Peak in Figure 3
linalyl acetate	34	31	40
geranial	35	40	41
thymol	36	51	42
carvacrol		52	43
methyl anthranilate	37	53	44
α-terpinyl acetate	38		45
neryl acetate	39	39	46
geranyl acetate	40	43	47
β-elemene			48
methyl N-methyl anthranilate	41	47	49
β-caryophyllene	42	34	50
α-humulene	43	37	51
methyl N-dimethyl anthranilate	44	48	52
α-selinene	45	41	53
bicyclogermacrene		42	54
(E,E)-α-farnesene	46		55
β-bisabolene			56
δ-cadinene	47	44	57
(Z)-3-hexenyl benzoate		50	58
spathulenol		49	59
caryophyllene oxide	48	46	60

The samples analyzed showed qualitative and quantitative differences even for some of the principal components, when they are compared with the data reported on the composition of Italian mandarin petitgrain oil (9). For example, our analyses showed the presence of p-cymene and also a higher content of α-thujene, limonene and γ-terpinene. Calvarano reported a concentration of methyl anthranilate of about 1%, while we found this compound as only a trace constituent.

Most of the oil components were found rather in narrow percentage ranges which supports the reliability of the isolation procedure of the oils. However, some quantitative variations among the different samples may be due both to the oil production technology and to the supply of the raw material for distillation. The leaves used for oil production were obtained from the pruning of *Citrus* trees, usually by unskilled workers. Different distillation conditions can cause variations of the content of some components: for example, the p-cymene content may vary when an oil undergoes different treatments. Also the amount of some alcohols may vary after treatment in a hot aqueous medium because they could have undergone the hydration/dehydratation reactions in equilibrium with monoterpene hydrocarbons.

Some other differences may be due to the presence of small quantities of different *Citrus* from other *Citrus* species, even after the careful selection of the leaves before distillation. For example, the higher amount (0.96%) of linalyl acetate in sample 4 may be due to the presence of 2% of bitter orange leaves, the oil of which contains about 30% of this compound (1). The higher content of sabinene (2.33%) and δ-3-carene (0.29%) of the sample 2 could be explained by the presence of about 3-4% of sweet orange leaves, the oil of which contains up to 50% of sabinene and up to 10% of δ-3-carene (24). However, the possibility of contamination of the raw material is limited, consequently the results obtained well characterize the composition of the Italian industrial mandarin petitgrain oil.

Table III. Quantitative composition (percentage) of mandarin petitgrain oil

Compounds*	1	2	3	4	5	Min	Max
tricyclene	t	t	t	t	t	t	t
α-thujene	0.97	0.96	0.78	0.98	1.04	0.78	1.04
α-pinene	2.11	2.12	1.75	2.16	2.30	1.75	2.30
α-fenchene	t	t	0.01	0.01	0.01	t	0.01
camphene	0.01	0.02	0.01	0.02	0.02	0.01	0.02
sabinene	0.90	2.33	0.22	0.30	0.30	0.22	2.33
β-pinene	2.09	2.13	1.90	2.31	2.45	1.90	2.45
6-methyl-5-hepten-2-one	t	t	t	0.01	0.01	t	0.01
myrcene	0.67	0.82	0.62	0.79	0.78	0.62	0.82
octanal	t	t	t	t	t	t	t
α-phellandrene	0.05	0.06	0.03	0.04	0.04	0.03	0.06
δ-3-carene	0.10	0.29	0.01	0.02	0.01	0.01	0.29
α-terpinene	0.33	0.32	0.19	0.24	0.26	0.19	0.33
o-cymene	t	t	t	t	t	t	t
p-cymene	2.96	3.26	4.84	5.19	4.54	2.96	5.19
limonene	7.18	7.76	9.41	12.59	11.65	7.18	12.59
β-phellandrene	0.05	0.04	0.03	0.04	0.04	0.03	0.05
1,8-cineole	0.01	0.01	0.02	0.02	0.02	0.01	0.02
(Z)-β-ocimene	0.17	0.17	0.15	0.18	0.20	0.15	0.20
(E)-β-ocimene	0.72	0.92	0.42	0.59	0.61	0.42	0.92
γ-terpinene	26.60	25.86	23.94	27.64	28.48	23.94	28.48
cis-sabinene hydrate	0.03	0.05	0.01	0.01	0.01	0.01	0.05
octanol	t	t	t	t	t	t	t
cis-linalool oxide (furanoid)	t	t	t	t	t	t	t
terpinolene	0.86	0.78	0.71	0.81	0.88	0.71	0.88
p-cymenene	0.13	0.15	0.10	0.16	0.18	0.10	0.18
linalool	0.64	1.10	0.27	0.93	0.29	0.27	1.10
nonanal	0.01	0.01	t	0.01	0.01	t	0.01
p-mentha-1,3,8-triene	0.01	t	0.02	0.01	0.02	t	0.02
cis-p-menth-2-en-1-ol	t	t	t	t	t	t	t
trans-p-menth-2-en-1-ol	0.03	0.02	0.02	0.02	0.02	0.02	0.03
citronellal	0.02	0.08	0.03	0.04	0.04	0.02	0.08
terpinen-4-ol	0.26	0.42	0.20	0.24	0.23	0.20	0.42
p-cymen-8-ol	0.01	0.01	0.01	0.02	0.01	0.01	0.02
α-terpineol	0.21	0.17	0.16	0.26	0.18	0.16	0.26
decanal	0.01	0.01	0.01	0.01	0.02	0.01	0.02
citronellol	t	t	t	t	t	t	t
nerol	0.01	0.03	0.02	0.03	0.02	0.01	0.03
methyl thymol	0.10	0.10	0.12	0.15	0.16	0.10	0.16
neral	t	t	0.01	0.06	0.03	t	0.06
geraniol	t	t	t	t	t	t	t
linalyl acetate	0.04	0.02	0.10	0.96	0.07	0.02	0.96
geranial	t	t	t	0.10	0.03	t	0.10
thymol	0.17	0.15	0.12	0.11	0.13	0.11	0.17
carvacrol	0.01	0.01	0.01	0.01	0.01	0.01	0.01
methyl anthranilate	t	0.01	0.03	0.03	t	t	0.03
α-terpinyl acetate	t	t	t	t	t	t	t
neryl acetate	t	0.02	0.02	0.05	0.04	t	0.05
geranyl acetate	t	0.01	t	t	0.02	t	0.02

Table III. Continued

Compounds*	1	2	3	4	5	Min	Max
β-elemene	t	t	t	t	0.01	t	0.01
methyl N-methyl anthranilate	50.62	47.88	51.93	41.61	43.19	41.61	51.93
β-caryophyllene	1.40	1.16	1.26	0.92	1.23	0.92	1.40
α-humulene	0.13	0.10	0.11	0.07	0.10	0.07	0.13
methyl N-dimethyl anthranilate	0.02	0.03	t	0.03	0.04	t	0.04
α-selinene	t	0.02	t	t	t	t	0.02
bicyclogermacrene	0.13	0.11	0.05	0.03	0.08	0.03	0.13
(E,E)-α-farnesene	0.03	0.02	t	t	t	t	0.03
β-bisabolene	t	t	t	0.02	0.01	t	0.02
δ-cadinene	t	t	t	t	t	t	t
(Z)-3-hexenyl benzoate	0.02	0.02	t	t	t	t	0.02
spathulenol	t	0.01	0.02	0.01	t	t	0.02
caryophyllene oxide	0.01	0.01	0.08	0.02	0.02	0.01	0.08
monoterpene hydrocarbons	45.91	47.99	45.14	54.08	53.81	45.14	54.08
sesquiterpene hydrocarbons	1.69	1.41	1.42	1.04	1.43	1.04	1.69
aldehydes	0.04	0.10	0.05	0.22	0.13	0.04	0.22
alcohols	1.37	1.97	0.84	1.64	0.90	0.84	1.97
esters	50.70	47.99	52.08	42.68	43.36	42.68	52.08
others	0.12	0.12	0.22	0.20	0.21	0.12	0.22

* = the compounds are listed according to the elution order on SE-52 column, 60 m; t = trace

References

1. L. Mondello, P. Dugo, G. Dugo and K. D. Bartle, *Italian Citrus petitgrain oils. Part I. Composition of bitter orange petitgrain oil.* J. Essent. Oil Res., **8**, 597-609 (1996).
2. L. Mondello, P. Dugo, A. Basile, G. Dugo and K. D. Bartle, *Interactive use of linear retention indices, on polar and apolar columns, with a MS-library for reliable identification of complex mixtures.* J. Microcol. Sep., **7**(6), 581-591 (1995).
3. B. M. Lawrence, *Progress in essential oils.* Perfum. Flavor., **18**(5), 43-68 (1993).
4. L. Peyron, *Petitgrain oils in perfumery.* Soap Perfum. Cosmet., **38**, 769-780 (1965).
5. L. Peyron, *Some little-known essential oils of potential interest in perfumery.* Soap Perfum. Cosmet., **39**, 633-643 (1966).
6. G. Vernin, *Détection et évaluation de l'anthranilate de méthyle et de ses dérives méthyles dans différents échantillons naturales et de synthèse par CCM et GLC.* La France et ses Parfums, **9**, 429-448 (1966).
7. J. A. Attaway, A. P. Pieringer and L. J. Barabas, *The origin of Citrus flavor components. I. The analysis of Citrus leaf oils using gas-liquid chromatography, thin layer chromatography and mass spectrometry.* Phytochemistry, **5**, 141-151 (1966).
8. J. A. Attaway, A. P. Pieringer and L. J. Barabas, *The origin of Citrus flavor components. III. A study of the percentage variations in peel and leaf oil terpenes during one season.* Phytochemistry, **6**, 25-32 (1967).
9. I. Calvarano, *Le essenze italiane di petitgrain. Nota I. I petitgrain di limone e mandarino.* Essenz. Deriv. Agrum., **37**, 27-54 (1967).
10. S. Kamiyama, *Studies on the leaf oils of Citrus species Part I. Composition of leaf oils from Citrus unshiu, Citrus natsudaidai, Citrus kokitsu, and Citrus limon.* Agric. Biol. Chem., **31**, 1091-1096 (1967).
11. S. Kamiyama, *Studies on the leaf oils of Citrus species. II. An examination of the seasonal variation of leaf oil composition.* Bull. Brew. Sci., **14**, 43-47 (1968).
12. S. Kamiyama and M. Amaha, *Studies on the leaf oil of Citrus species. VI. Composition of leaf oils from ten Citrus taxa and some intrageneric hybrids.* Bull. Brew. Sci., **18**, 17-27 (1972).
13. A. Di Giacomo, *Gli olii essenziali degli agrumi.* Rivista Ital. EPPOS, 249-256, Milano (1974).

14. R. W. Scora, A. E. Esen and J. Kumamoto, *Distribution of essential oils in leaf tissue of an F_2 population of Citrus*. Euphytica, **25**, 201-209 (1976).
15. L. R. de Vottero, G. Tonarelli, E. R. de Krasnogor and J. A. Retamar, *Petitgrains de hojas de naranjo amargo, limonero y mandarinero*. Essenz. Deriv. Agrum., **2**, 150-156 (1978).
16. M. S. Karawya and M. S. Hifnawy, *Leaf essential oils of three different varieties of Citrus reticulata Blanco growing in Egypt*. Perfum. Flavor., **4**(2), 27-30 (1979).
17. J. M. Ortiz Marcide, J. L. Tadeo Lluch, F. J. Diaz Llanos, A. Estellas Adam, *Etude des huiles essentielles de feuilles du groupe de la mandarine clementine. Utilisation taxonomique*. Fruits, **38**, 125-131 (1983).
18. N. A. Kekelidze, M. I. Janikashvili and A. P. Kachurina, *Accumulation of mono- and sesquiterpenes in mandarin leaves and fruits of tangerine in ontogenesis*. Fiziol. Biokhim. Kul't. Rast., **18**, 287-292 (1986).
19. O. Ekundayo, O. Bakare, A. Adesomoju and E. Stahl-Biskup, *Leaf volatile oil composition of mandarin (Citrus reticulata) from Nigeria*. J. Essent. Oil Res., **2**, 329-330 (1990).
20. Z. Fleisher and A. Fleisher, *Mandarin leaf oil (Citrus reticulata Blanco). Aromatic plants of the Holy Land and the Sinai. Part III*. J. Essent. Oil Res., **2**, 331-334 (1990).
21. K. Ogihara, K. Munesada and T. Suga, *Variations in leaf terpenoids with ploidy level in Citrus cultivars and hybrids*. Phytochemistry, **29**, 1889-1891 (1990).
22. Z. K. Lin and Y. F. Hua, *Systematic evolutional relation of chemical components of the essential oil from 11 taxa of Citrus leaves*. Acta Bot. Sinica, **34**, 133-139 (1992).
23. C. Blanco Tiredo, E. E. Stashenko, M. Y. Combariza and J. R. Martinez, *Comparative study of Colombian Citrus oils by high-resolution gas chromatography and gas chromatography-mass spectrometry*. J. Chromatogr., **697**, 501-513 (1995).
24. L. Mondello, A. Cotroneo, I. Stagno d'Alcontres and G. Dugo, *Italian Citrus petitgrain oils. Part III. Composition of sweet orange petitgrain oil*. J. Essent. Oil Res., in press (1997).