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Short communication

Gas chromatographic–mass spectrometric analysis of essential oils from *Pimpinella aurea*, *Pimpinella corymbosa*, *Pimpinella peregrina* and *Pimpinella puberula* gathered from Eastern and Southern Turkey

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Abstract

Essential oils from fruits, stems and leaves and roots of *Pimpinella aurea* DC., *P. corymbosa* Boiss., *P. peregrina* L. were analyzed by gas chromatography (GC) and gas chromatography–mass spectrometry (GC–MS) techniques. Fruits and aerial parts of *P. puberula* (DC.) Boiss were also evaluated. A total of 140 different compounds were identified, and significant qualitative and quantitative differences were observed among the samples. In fact, the main constituents of each species were different and only the oils extracted from roots shared the same principal compound, epoxy pseudoisoeugenyl-2-methyl butyrate (26.8–42.8%). The other fractions were dominated by different sesquiterpene compounds although in three of them, *P. aurea* stem and leaves, *P. puberula* fruits and *P. puberula* stems and leaves, monoterpene constituents also appear as main ones.

Keywords: *Pimpinella aurea*; *Pimpinella corymbosa*; *Pimpinella peregrina*; *Pimpinella puberula*; Essential oils; GC; GC–MS; Phenylpropanoids; Sesquiterpenes; Monoterpenes

1. Introduction

Pimpinella is a member of the Apiaceae and comprises approximately 150 species distributed in the northern hemisphere [1]. *Pimpinella* is represented in Turkey by 23 species (5 endemic), 2 subspecies and 2 varieties, representing a total of 27 [2]. Both extracts and essential oils of *Pimpinella* species are known to have high content of phenylpropanoid derivatives. The 2-hydroxy-5-methoxy-1-(*E*)-propenylbenzene skeleton of these compounds, known as pseudoisoeugenol, is unique to *Pimpinella* [3]. One important criterium for studying the oils of this genus is the fact that they contain C_{12} sesquiterpenoids and phenylpropanoids with a unique structure and their biological activities may have potential for developing new agents for use in agriculture and medicine [3–6]. These phenylpropanoids show various bioactivities such as antigermination, insecticidal, acaricidal, weak antitumor, antimalarial, antimicrobial and antifungal [4–9]. The essential oils from different plant parts of *Pimpinella aurea* grown in Iran were previously reported [10,11]. Our earlier investigations of *Pimpinella* species resulted in the isolation of four new and eighteen known compounds [4–6]. The chemical composition, genetic diversity and activity of essential oils of several *Pimpinella* species were previously reported in our studies [5,7]. Here, we report on detailed analysis of essential oils from different parts of *Pimpinella corymbosa*, *Pimpinella peregrina*, and *Pimpinella puberula* by GC and GC–MS.

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2. Experimental

2.1. Plant material

The samples of each *Pimpinella* species were collected from Eastern and Southern Turkey in June/July 2001. Collection localities, dates, plant parts, and essential oil yield are given in Table 1. Voucher specimens were placed at the Herbarium of the Faculty of Pharmacy, Anadolu University in Eskisehir, Turkey (ESSE). Botanical identifications were carried out by Professor Dr. Zeki Aytac, Gazi University, Ankara, Turkey.

2.2. Isolation of essential oils

Air-dried fruits, stems and leaves and roots were crushed separately using a mortar followed by water distillation for 3 h using a modified Clevenger-type apparatus to obtain essential oils [12]. Percent yield of oils calculated on a moisture free basis are shown in Table 1.

2.3. Gas chromatography

Essential oils were analyzed by GC using a Hewlett Packard 6890 system. An HP-Innowax FSC (60 m × 0.25 mm I.D., with 0.25 μm film thickness) was used with nitrogen as the carrier gas (1 mL/min). The oven temperature was kept at 60 °C for 10 min, programmed to reach 220 °C at a rate of 4 °C/min, then kept constant at 220 °C for 10 min before proceeding to 240 °C at a rate of 1 °C/min. The split ratio was adjusted to 50:1. Flame ionization detection and injector temperature were performed at 250 °C.

2.4. Gas chromatography–mass spectrometry

Essential oils were analyzed by GC–MS using a Hewlett-Packard GCD system. Innowax FSC column (60 m × 0.25 mm, 0.25 μm film thickness) was used with helium as the carrier gas (1 mL/min). GC oven temperature and conditions were as described above. The injector temperature was at 250 °C. Mass spectra were recorded at 70 eV. Mass range was from *m*/*z* 35 to 425.

2.5. Identification of essential oil constituents

The quantification of the components was performed on the basis of their GC peak areas on the Innowax column and percentages of the characterized essential oil components were as given in Table 2. The identification of the separated volatile organic compounds was achieved through retention indices and mass spectrometry by the comparing mass spectra of the unknown peaks with those stored in the Wiley GC/MS Library, MassFinder and the in-house "Baser Library of Essential Oil Constituents" which includes over 3200 genuine compounds with MS and retention data. *n*-Alkanes (C9–C20) were used as reference points in the calculation of retention indices (I) [13–15]. After the GC/MS analysis, the constituents **60**, **89**, **97**, **107**, **112**, **121**, **125**, **128**, **129**, **131**, **132**, **133**, **135**, **136**, **137**, **139** were not identifiable in the Wiley GC/MS Library and the Baser Library of Essential Constituents. Separation of these compounds was, therefore, necessary. Essential oils of *Pimpinella* species were subjected to column chromatography (silica gel) using *n*-hexane and diethyl ether according to our previous procedure [4–6]. Structure elucidation of the isolated compounds was achieved by a combination of one-dimensional (1D) and two-dimensional (2D) NMR techniques on Bruker Avance DRX 500 at 500 (^{1}H) and 125 MHz (^{13}C), Bruker DRX 400 at 400 (^{1}H) and 100 MHz (^{13}C) and Bruker DRX 300 at 300 (^{1}H) and 75 MHz (^{13}C) instruments, LC-electrospray ionization (ESI) MS, ESI-MS and the known compounds were compared with literature values [8,16–20]. Isolated compounds were re-analyzed by GC–MS to confirm their identity with the constituents of the essential oils. Their mass spectral fragmentation patterns are given in Table 3.

3. Results and discussion

Essential oil yields of the different parts of *P. aurea*, *P. corymbosa*, *P. peregrina* and *P. puberula* were variable and ranged from 0.3 to 5.1%; 0.1 to 0.3% and 0.1 to 0.2% (Table 1). Essential oils

Table 1
Plant materials used in this study

Species	Collection site	Collection date	Plant part	Code	Yield[a] (%)	ESSE[b]
P. aurea DC.	Van: Van-Ercis road	July 2001	Fruits	PaF	5.1	13912
			Stems and leaves	PaSL	0.3	
			Roots	PaR	0.1	
P. corymbosa Boiss.	Siirt: Siirt-Sirnak road	July 2001	Fruits	PcF	0.3	13899
			Stems and leaves	PcSL	0.2	
			Roots	PcR	0.2	
P. peregrina L.	Antalya: Akseki, Guzelsu-Serebil,	June 2001	Fruits	PpeF	1.1	13877
			Stems and leaves	PpeSL	0.1	
			Roots	PpeR	0.1	
P. puberula (DC.) Boiss.	Hakkari: Hakkari-Van road	July 2001	Fruits	PpuF	0.3	13909
			Stems and leaves	PpuSL	0.2	

Pa: *Pimpinella aurea*; Pc: *Pimpinella corymbosa*; Ppe: *Pimpinella peregrina*; Ppu: *Pimpinella puberula*; F: fruits; SL: stems and leaves; R: roots.
[a] Calculated on moisture-free basis.
[b] Herbarium of the Faculty of Pharmacy, Anadolu University in Eskisehir, Turkey (ESSE).

Table 2
Essential oil composition (%) of different tissues of *Pimpinella aurea*, *P. corymbosa*, *P. peregrina* and *P. puberula*

No.	Compound	I[a]	PaF	PaSL	PaR	PcF	PcSL	PcR	PpeF	PpeSL	PpeR	PpuF	PpuSL
1	α-Pinene	1032	1.4	12.1	3.6	0.6	0.2	–	0.1	0.5	–	0.1	tr
2	Camphene	1076	tr	0.2	–	0.3	0.1	–	–	–	–	–	–
3	β-Pinene	1118	1.9	8.0	1.7	2.0	1.1	–	–	–	–	0.1	0.1
4	Sabinene	1132	4.3	20.7	3.1	0.1	0.1	–	–	–	–	5.4	3.3
5	δ-3-Carene	1159	0.1	0.8	0.5	–	–	–	–	–	–	–	–
6	Myrcene	1174	0.1	0.8	0.2	0.2	0.2	–	–	–	–	0.5	0.3
7	α-Terpinene	1188	0.1	0.4	0.1	–	–	–	–	–	–	0.1	0.1
8	Limonene	1203	0.1	1.2	0.3	0.2	0.1	–	–	–	–	63.4	36.5
9	β-Phellandrene	1218	0.1	0.6	0.2	–	–	–	–	–	–	0.1	tr
10	(*Z*)-3-Hexanal	1225	–	–	–	–	0.1	–	–	–	–	0.1	0.1
11	γ-Terpinene	1255	0.2	0.8	0.2	–	tr	tr	–	–	–	–	–
12	(*E*)-β-Ocimene	1266	0.1	0.1	–	–	–	–	–	–	–	–	–
13	*p*-Cymene	1280	0.1	0.4	0.1	–	0.1	tr	–	–	–	tr	tr
14	Terpinolene	1290	0.1	0.3	0.1	–	–	–	–	–	–	tr	tr
15	Octanal	1296	–	–	0.1	–	–	–	–	–	–	–	–
16	Isogeijerene	1304	–	–	–	–	–	0.5	–	–	–	0.3	0.9
17	Geijerene	1338	0.1	tr	0.1	–	0.1	4.4	0.1	–	0.1	2.4	7.3
18	6-Methyl-5-hepten-2-one	1348	–	tr	–	0.1	tr	–	–	–	–	–	–
19	Hexanol	1360	–	–	–	0.2	tr	–	–	–	–	–	–
20	Nonanal	1400	–	tr	–	0.2	tr	–	–	–	–	–	–
21	α,*p*-Dimethylstyrene	1452	–	tr	–	–	–	–	–	–	–	–	–
22	Clavukerin B	1455	–	–	–	–	–	0.2	–	–	–	–	–
23	*cis*-1,2-Limonene epoxide	1458	–	–	–	–	–	–	–	–	–	–	tr
24	α-Cubebene	1466	–	–	–	0.2	0.2	–	–	–	–	–	–
25	*trans*-Sabinene hydrate	1474	0.1	0.3	0.1	–	–	–	–	–	–	tr	tr
26	δ-Elemene	1479	–	–	–	0.3	0.3	tr	–	–	–	–	–
27	Isogeijerene C	1493	–	–	–	–	–	–	–	–	–	tr	0.2
28	Bicycloelemene	1495	0.1	0.1	tr	0.2	0.2	tr	–	–	–	–	–
29	α-Copaene	1497	0.2	0.1	0.1	4.5	4.0	0.2	0.1	0.3	0.1	–	–
30	β-Bourbonene	1535	tr	tr	–	0.2	0.1	–	–	–	–	–	–
31	Benzaldehyde	1541	–	tr	–	–	–	–	–	–	–	–	–
32	α-Bergamotene	1545	–	–	–	–	–	–	–	–	2.7	–	–
33	(*E*)-2-Nonenal	1548	–	–	0.1	–	–	–	–	–	–	–	–
34	β-Cubebene	1549	–	tr	–	1.2	1.0	0.1	–	–	–	–	–
35	*cis*-Sabinene hydrate	1556	tr	0.2	–	–	–	–	–	–	–	–	–
36	*trans*-*p*-Menth-2-en-1-ol	1571	–	0.1	–	–	–	–	–	–	–	–	–
37	Pinocarvone	1586	–	–	0.1	–	–	–	–	–	–	–	–
38	Isocaryophyllene	1589	–	–	–	–	–	–	0.1	0.2	–	–	–
39	*trans*-β-Bergamotene	1594	–	–	–	–	–	–	70.3	40.9	–	–	–
40	Pregeijerene	1594	–	–	–	0.1	0.2	1.1	–	–	–	0.6	1.9
41	Bornyl acetate	1597	0.1	0.3	0.1	0.8	0.8	tr	–	–	–	–	–
42	β-Elemene	1600	tr	0.1	–	0.3	–	–	–	–	–	–	–
43	Terpinen-4-ol	1611	0.1	0.1	0.2	–	–	–	–	–	–	tr	tr
44	β-Caryophyllene	1612	0.4	2.0	0.2	33.2	32.5	0.3	4.7	5.1	0.3	0.2	0.4
45	*cis*-*p*-Menth-2-en-1-ol	1638	–	0.1	–	–	–	–	–	–	–	–	–
46	*trans*-*p*-Mentha-2,8-dien-1-ol	1639	–	–	–	–	–	–	–	–	–	–	0.1
47	Myrtenal	1648	–	tr	0.1	–	–	–	–	–	–	–	–
48	9-*epi*-β-Caryophyllene	1655	–	–	–	–	–	–	–	–	0.2	–	–
49	α-Himachalene	1661	–	–	–	–	0.1	0.1	–	–	–	–	–
50	(*Z*)-β-Farnesene	1668	0.1	0.2	0.1	0.2	0.2	tr	0.9	0.4	–	–	–
51	*trans*-Pinocarveol	1670	tr	tr	0.2	–	–	–	–	–	–	–	–
52	*cis*-*p*-Mentha-2,8-dien-1-ol	1678	–	–	–	–	–	–	–	–	–	–	0.1
53	*trans*-Verbenol	1683	–	0.1	0.2	–	–	–	–	–	–	–	–
54	α-Humulene	1687	–	–	–	7.1	7.3	0.1	1.2	1.5	–	tr	0.1
55	(*E*)-β-Farnesene	1695	–	–	–	–	–	–	0.1	0.1	–	–	–
56	Guaioxide	1697	1.3	1.1	0.4	–	–	–	–	–	–	–	–
57	*p*-Mentha-1,8-dien-4-ol	1700	0.1	0.2	–	–	–	–	–	–	–	–	–
58	γ-Muurolene	1704	0.7	0.3	0.3	0.4	0.8	0.1	0.1	0.1	–	–	–
59	γ-Himachalene	1705	–	–	–	0.8	0.7	0.4	–	–	–	–	–
60	4,6-Guaiadiene	1714	–	–	–	0.6	–	–	–	0.1	–	–	–
61	Dodecanal	1722	–	–	tr	–	–	–	–	–	–	–	–
62	Germacrene D	1722	5.7	2.9	3.7	9.1	11.6	0.8	0.3	0.8	–	0.1	0.2
63	α-Zingiberene	1725	–	–	–	–	–	0.2	–	–	–	–	–

Table 2 (*Continued*)

No.	Compound	I[a]	PaF	PaSL	PaR	PcF	PcSL	PcR	PpeF	PpeSL	PpeR	PpuF	PpuSL
64	7-*epi*-1,2-Dehydrosesquicineole	1727	–	–	–	2.4	–	–	–	–	–	–	–
65	*cis*-α-Bergamotene	1740	–	–	–	–	–	–	–	–	0.2	–	–
66	β-Bisabolene	1741	33.1	9.6	6.1	2.4	–	2.5	1.2	1.3	0.8	–	0.1
67	α-Cadinene	1743	–	–	–	1.0	–	–	–	–	–	–	–
68	Carvone	1751	–	–	–	–	–	–	–	–	–	tr	0.1
69	Bicyclogermacrene	1755	0.2	–	0.2	2.2	3.9	1.0	0.9	0.5	–	–	–
70	Geranyl acetate	1765	–	–	–	–	0.1	–	–	–	–	–	–
71	δ-Cadinene	1772	–	–	–	1.5	1.7	–	–	–	–	–	–
72	β-Sesquiphellandrene	1783	–	–	–	–	–	–	0.4	0.6	9.5	–	–
73	(*E*)-α-Bisabolene	1784	–	–	–	0.2	0.2	–	–	–	–	–	–
74	Kessane	1786	2.1	1.8	0.7	–	–	–	–	–	–	–	–
75	3,10-Dihydro-1,4-dimethylazulene	1786	–	–	–	–	–	–	–	–	–	–	tr
76	*p*-Methyl acetophenone	1797	–	–	–	–	–	–	–	–	–	–	tr
77	Cadina-1,4-diene	1799	–	–	–	0.1	0.1	–	–	–	–	–	–
78	Myrtenol	1804	tr	0.1	0.1	–	–	–	–	–	–	–	–
79	Perilla aldehyde	1807	–	–	–	–	–	–	–	–	–	–	tr
80	4,10-Dihydro 1,4-dimethylazulene	1815	–	–	–	–	–	1.3	–	–	–	–	–
81	(*E,E)*-2,4-Decadienal	1827	–	–	–	–	–	0.1	–	–	–	–	–
82	(*E*)-β-Damascenone	1838	–	–	–	–	tr	–	–	–	–	–	0.1
83	(*E*)-Anethole	1845	0.1	0.2	0.2	0.1	0.1	0.1	0.3	0.4	0.3	–	–
84	*trans*-Carveol	1845	–	–	–	–	–	–	–	–	–	–	tr
85	*p*-Cymen-8-ol	1864	–	0.1	–	–	–	–	–	–	–	–	–
86	Dehydrocostus lactone	1867	–	–	–	–	–	–	–	–	–	0.2	1.3
87	(*E*)-Geranyl acetone	1868	–	0.1	–	–	0.1	–	–	–	–	–	–
88	Benzyl-2-methylbutyrate	1880	–	–	–	–	0.1	–	–	–	–	–	–
89	Traginone = 4-(6-methyl-bicyclo[4.1.0]hept-2-en-7-yl)-butan-2-one	1881	–	–	–	–	–	–	–	–	–	0.2	1.2
90	*epi*-Cubebol	1900	–	–	–	0.1	0.1	–	–	–	–	–	–
91	α-Calacorene	1941	–	–	–	–	tr	–	–	–	–	–	–
92	4-Hydroxy-2-methyl acetophenone	1942	–	–	–	0.7	0.9	tr	–	–	–	–	–
93	Dodecanol	1973	–	–	0.2	–	–	–	–	–	–	–	–
94	Isocaryophyllene oxide	2001	–	–	–	0.4	0.4	–	tr	0.4	–	–	–
95	Caryophyllene oxide	2008	0.1	0.4	0.5	3.1	3.5	0.2	0.5	2.9	–	0.1	0.5
96	Methyl eugenol	2030	–	–	–	–	0.1	–	–	–	–	23.1	29.6
97	Aurean = 1-methyl-4-(5-methyl-1-methylene-hex-4-enyl)-7-oxa-bicyclo [4.1.0] heptane	2038	33.5	19.8	9.7	–	–	–	1.2	2.7	0.2	–	–
98	2-Methyl-5-methoxy-benzofuran	2060	–	–	–	–	–	–	0.1	0.4	–	–	–
99	α-Copaene-11-ol	2065	–	–	–	2.5	2.5	0.1	–	–	–	–	–
100	Humulene epoxide II	2071	–	–	–	–	–	–	0.1	0.8	0.1	tr	0.2
101	1-*epi*-Cubenol	2088	–	–	–	0.2	–	–	–	–	–	–	–
102	*p*-Cresol	2094	1.4	0.1	–	1.8	1.1	0.1	–	–	–	–	–
103	Elemol	2096	–	–	–	–	–	–	–	–	–	–	0.1
104	Spathulenol	2144	0.1	0.4	0.2	0.7	0.8	0.7	–	–	–	–	–
105	8-*epi*-Dictamnol	2152	–	–	–	–	–	–	–	–	–	–	0.1
106	β-Bisabolenol	2158	0.1	0.2	0.5	–	–	–	–	–	–	–	–
107	Dictamnol	2170	–	–	–	–	–	–	–	–	–	0.6	2.8
108	4-(1-Propenyl)-phenyl isobutyrate	2182	–	–	–	–	–	0.5	–	–	–	–	–
109	Eugenol	2186	–	–	–	–	–	–	–	–	–	–	tr
110	T-Cadinol	2187	–	–	–	0.3	0.3	–	–	–	–	–	–
111	T-Muurolol	2209	–	–	–	0.1	0.1	tr	–	–	–	–	–
112	*trans*-Isoosmorhizole	2212	–	–	–	–	–	–	0.1	0.3	0.1	–	–
113	α-Bisabolol	2232	–	0.3	–	0.1	0.1	–	0.7	3.1	0.4	–	–
114	Carvacrol	2239	–	0.3	1.2	0.2	0.2	–	0.1	0.1	0.1	–	tr
115	Himachalol	2240	–	–	–	0.4	–	–	–	–	–	–	–
116	Elemicine	2245	–	–	–	–	–	–	–	–	–	tr	0.1
117	*trans*-α-Bergamotol	2247	–	–	–	–	0.7	0.2	–	–	–	–	–

Table 2 (*Continued*)

No.	Compound	I[a]	PaF	PaSL	PaR	PcF	PcSL	PcR	PpeF	PpeSL	PpeR	PpuF	PpuSL
118	α-Cadinol	2255	–	–	–	0.1	–	–	–	–	–	–	–
119	Sinensal	2262	–	–	–	–	–	–	0.3	–	–	–	–
120	*cis*-Guaia-9-en-11-ol	2265	–	–	–	0.2	–	–	–	–	–	–	–
121	4-(1-Propenyl)-phenyl-2-methyl butyrate	2284	1.6	1.1	1.4	–	0.4	33.8	0.3	2.1	0.1	–	–
122	1,4-Dimethyl azulene	2291	–	–	–	–	–	1.0	–	–	–	–	0.2
123	(*Z*)-α-*trans*-β-Bergamotol acetate	2296	–	–	–	0.9	0.6	tr	–	–	–	–	–
124	Caryophylladienol II	2324	–	–	–	–	–	–	–	0.2	0.1	–	–
125	12-Hydroxy-β-caryophyllene acetate	2331	–	0.2	–	4.9	3.1	tr	–	–	–	–	–
126	Caryophyllenol II	2392	–	–	–	–	–	–	–	–	–	–	tr
127	Eudesma-4 (15),7-dien-1β-ol	2370	–	0.3	0.5	–	–	–	–	–	–	–	–
128	4-(1-Propenyl)-phenyl tiglate	2406	2.5	1.2	1.6	–	–	–	–	–	–	–	–
129	4-Methoxy-2-(1-propenyl)-phenyl isobutyrate	2462	–	–	–	–	–	–	tr	0.3	0.5	–	–
130	Dodecanoic acid	2503	–	–	0.2	–	–	–	–	–	–	–	–
131	4-(3-Methyloxiranyl)-phenyl-2-methyl butyrate	2506	1.2	0.4	1.9	–	–	–	–	–	0.3	–	–
132	Pseudoisoeugenyl-2-methyl butyrate	2567	–	0.1	4.4	–	–	–	–	–	0.8	–	–
133	4-Methoxy-2-(3-methyloxiranyl)-phenyl isobutyrate	2613	–	–	–	–	–	–	3.7	5.5	44.8	–	–
134	Sedanolide	2618	–	–	–	–	–	–	–	–	–	–	0.1
135	4-(3-Methyloxiranyl)-phenyl tiglate	2642	0.3	–	–	–	–	–	–	–	–	–	–
136	Epoxy pseudoisoeugenyl-2 methyl butyrate	2698	–	–	39.0	–	0.1	42.8	0.4	0.5	26.8	–	0.1
137	4-Methoxy-2-(3-methyloxiranyl)-phenyl angelate	2825	–	–	–	–	–	–	–	8.1	0.1	–	–
138	Pentadecanoic acid	2922	–	–	0.2	–	–	–	–	–	–	–	–
139	4-Methoxy-2-(3-methyloxiranyl)-phenyl tiglate	2926	–	–	0.2	–	–	–	–	0.6	tr	–	–
140	Hexadecanoic acid	2931	0.1	0.3	1.2	0.2	0.1	0.9	0.8	1.6	1.0	0.2	2.3
	Total identified (%)		94.0	91.6	86.4	89.9	83.4	93.8	89.1	82.4	89.6	97.8	90.5

Pa: *Pimpinella aurea*; Pc: *Pimpinella corymbosa*; Ppe: *Pimpinella peregrina*; Ppu: *Pimpinella puberula;* F: fruits; SL: stems and leaves; R: roots; tr: trace (≤0.1%).
[a] Retention indices on an innowax column calculated against *n*-alkanes (C9–C20).

were analyzed by GC and GC–MS systems using a polar column and the reliable in-house Baser Library of Essential Oil Constituents which contain mass spectra and retention data on over 3200 genuine compounds. One hundred and forty compounds were identified in *Pimpinella* essential oils obtained from different parts constituting 82.4 to 97.8% of the total oil. These compounds are listed in Table 2 with their relative percentages.

The *P. aurea* fruit oil (PaF) was characterized by a high content of a new epoxybisabolol-type oxygenated sesquiterpene; 1-methyl-4-(5-methyl-1-methylene-hex-4-enyl)-7-oxa-bicyclo[4.1.0]heptane trivially named as aurean (33.5%) and β-bisabolene (33.1%) (Table 2). In our previous study, we isolated and identified aurean from *P. aurea* fruit oil [4]. Oil from *P. aurea* stems and leaves (PaSL) yielded sabinene (20.7%), aurean (19.8%), α-pinene (12.1%) and β-bisabolene (9.6%) as major components. The phenylpropanoid, epoxypseudoisoeugenol-2-methylbutyrate, EPB, (39.0%), was detected as unique main component in the root oil (PaR). Besides the EPB, aurean (9.7%) and β-bisabolene (6.1%) were also identified as the other most abundant compounds. The chemical composition of essential oils from different parts of *P. aurea* collected from different localities in Iran has previously been reported [10,11]. According to Assadian et al. [10], aerial parts *P. aurea* collected from Northern Tehran was characterized by high amounts of *trans*-α-bergamotene (72.8%). The major constituents of *P. aurea* from stem/leaves (SL), flowers (F), and seed (S) obtained from Northwestern Tehran [11] were α-pinene (SL: 13.0%; F: 1.6%; S: 0%), limonene +1,8-cineole (SL: 21.0%; F: 9.0%), viridiflorol (SL: 13.0%; F: 33.0%; S: 37.0%) and β-bisabolene (SL: 4.0%; F: 30.0%; S: 51.0%). Viridiflorol, which

Table 3
Mass spectral fragmentation patterns of the isolated compounds

No.[a]	I[b]	Mass spectral fragmentation
60	1714	($C_{15}H_{24}$) 204 [M^+] (59), 189 (58), 175 (5), 162 (14), 161 (79), 147 (27), 133 (39), 121 (16), 119 (68), 107 (32), 105 (93), 95 (19), 93 (35), 91 (88), 81 (32), 79 (40), 77 (45), 67 (23), 66 (27), 65 (23), 55 (46), 53 (28), 43 (37), 42 (20), 41 (100)
89	1881	($C_{12}H_{18}O$) 178 [M^+] (2), 161 (5), 145 (9), 131 (3), 120 (35), 119 (14), 118 (14), 107 (13), 105 (72), 93 (26), 92 (35), 91 (65), 79 (59), 77 (36), 65 (16), 55 (19), 53 (15), 43 (100), 41 (34)
97	2038	($C_{15}H_{24}O$) 220 [M^+] (4), 177 (4), 159 (8), 131 (8), 119 (10), 109 (25), 93 (25), 91 (29), 79 (30), 77 (22), 69 (100), 67 (35), 55 (32), 53 (27) 51 (7)
107	2170	($C_{12}H_{18}O$) 178 [M^+] (2), 163 (6), 160 (15), 145 (27), 131 (19), 120 (32), 119 (15), 118 (18), 117 (26), 115 (14), 107 (16), 105 (63), 93 (32), 92 (37), 91 (75), 85 (3), 79 (54), 77 (21), 71 (39), 65 (21), 55 (19), 53 (21), 43 (100), 41 (53)
112	2212	($C_{11}H_{14}O_2$) 178 [M^+] (100), 177 (24), 163 (23), 151 (11), 149 (32), 147 (26), 135 (19), 131 (9), 121 (43), 115 (8), 105 (19), 103 (40), 91 (37), 79 (14), 77 (37), 69 (9), 65 (16), 51 (21), 41 (9)
121	2284	($C_{14}H_{18}O_2$) 218 [M^+] (4), 135 (10), 134 (100), 133 (38), 105 (8), 91 (6), 79 (9), 77 (17), 65 (4), 57 (56), 56 (7), 51 (6)
125	2331	($C_{17}H_{26}0_2$) 262 [M^+] (5), 234 (5), 220 (5), 202 (8), 189 (19), 187 (22), 174 (8), 159 (31), 145 (23), 133 (42), 119 (46), 107 (42), 105 (57), 93 (59), 91 (63), 79 (45), 77 (35), 67 (30), 55 (23), 53 (27), 41 (22), 43 (100)
128	2406	($C_{14}H_{16}O_2$) 216 [M^+] (10), 134 (26), 133 (8), 135 (3), 103 (3), 84 (5), 43 (89), 79 (5), 78 (4), 77 (9), 65 (3), 56 (4), 55 (100), 53 (8), 51 (6)
129	2462	($C_{13}H_{16}O_2$) 204 [M^+] (7)161 (2), 145 (10), 135 (12), 134 (100), 133 (41), 119 (5), 105 (12), 103 (12), 93 (11), 91 (11), 79 (14), 77 (17), 71 (11), 55 (11), 43 (56), 41 (26).
131	2506	($C_{14}H_{18}O_3$) 234 [M^+] (2), 150 (13), 149 (9), 133 (9), 121 (7), 107 (15), 106 (26), 105 (9), 85 (22), 84 (18), 78 (14), 77 (21), 57 (100), 51 (20), 43 (8), 41 (25)
132	2567	($C_{15}H_{20}O_3$) 248 [M^+] (5), 165 (12), 164 (100), 149 (23), 135 (3), 121 (3), 105 (5), 91 (8), 77 (6), 66 (5), 55 (3), 57 (32), 41 (14)
133	2613	($C_{14}H_{18}O_4$) 250 [M^+] (20), 180 (32), 163 (44), 162 (39), 151 (32), 138 (21), 137 (79), 135 (20), 123 (6), 105 (5), 92 (16), 77 (15), 71 (16), 65 (11), 51 (7), 43 (100), 41 (23)
135	2642	($C_{14}H_{16}O_3$), 232 [M^+] (3), 107(14), 84 (6), 83 (100), 77 (7), 69 (9), 55 (75), 53 (7), 43 (25), 41 (8)
136	2698	($C_{15}H_{20}O_4$) 264 [M^+] (5), 180 (41), 161 (25), 162 (43), 151 (33), 137 (43), 119 (12), 105 (5), 91 (17), 77 (17), 65 (15), 57 (100), 56 (10)
137	2825	($C_{15}H_{18}O_4$) 262 [M^+] (4), 180 (5), 164 (6), 163 (12), 162 (36), 151 (4), 147 (9), 137 (6), 119 (5), 91 (8), 84 (5), 83 (92), 77 (7), 65 (8), 63 (5), 56 (5), 55 (100), 54 (8), 53 (9), 51 (6)
139	2926	($C_{15}H_{18}O_4$) 262 [M^+] (3), 246 (6), 180 (4), 164 (14), 163 (8), 162 (35), 147 (12), 137 (4), 119 (7), 103 (4), 91 (13), 83 (100), 84 (5), 77 (9), 65 (11), 63 (5), 55 (97), 54 (8), 53 (13), 51 (87)

[a] The numbers in bold represent the compounds in Table 2.
[b] I: retention indices on an innowax column calculated against *n*-alkanes (C9–C20).

was identified in sizable amounts in the stem/leaves obtained from the Northwestern Tehran samples [11], was not found in the oil from the aerial parts of *P. aurea* (PaA) investigated in our work. This is not unexpected by the fact that plants often produce different amounts of phytochemicals when grown in different geographical locations.

Composition of *P. corymbosa* fruits (PcF) or stems and leaves (PcSL) were dominated by sesquiterpenes such as β-caryophyllene (33.2 and 32.5%), germacrene D (9.1 and 11.6%) and α-humulene (7.1 and 7.3%) (Table 2). The main compounds of the root oil (PcR) were epoxypseudoisoeugenol-2-methylbutyrate (42.8%) and 4-(1-propenyl)-phenyl-2-methylbutyrate (4-PM) (33.8%). The latter 4-PM-compound was isolated from *Moonia heterophylla* (Compositae) [21]. We have previously isolated it from *P. corymbosa* root oil [4], being the first report of this compound in *Pimpinella* species.

trans-α-Bergamotene was the major compound obtained from *P. peregrina* fruits (PpeF) (70.3%) and stems and leaves (PpeSL) (40.9%) whereas the root oil (PpeR) contained 4-methoxy-2-(3-methyloxiranyl)-phenylisobutyrate (44.8%) and epoxypseudoisoeugenol-2-methylbutyrate (26.8%) as major compounds (Table 2). The 4-methoxy-2-(3-methyloxiranyl)-phenylisobutyrate was isolated for the first time from *P. saxifraga* [22] and later it has also been identified in other species of this genus such as *P. anisum*, *P. aromatica*, *P. cumbrae*, *P. diversifolia*, *P. junoniae*, *P. major*, *P. nigra*, *P. peregrina*, *P. tripartrita*, *P. villosa* and *P. isaurica* [3,5,23,25–28].

The essential oil profile of *P. puberula* fruits (PpuF) and stems and leaves (PpuSL) was significanlty different from those of other *Pimpinella* species, with high proportion of monoterpenes such as limonene (63.4–36.5%) and methyl eugenol (23.1–29.6%) in the oils (Table 2). *P. puberula* essential oil profile also consisted of a variety of characteristic C_{12} sesquiterpenes (geijerenes and azulenes) in low concentrations: geijerene (2.4–7.3%), isogeijerene (0.3–0.9%), isogeijerene C (0.03–0.2%), pregeijerene (0.6–1.9%), 3,10-dihydro-1,4-dimethylazulene (0.03% to trace) and 1,4-dimethylazulene (0.2% to trace). We were unable to obtain the essential oil from *P. puberula* roots due to insufficient plant material.

The occurrence of rare phenylpropanoids from pseudoisoeugenol group, such as epoxy pseudoisoeugenol-2-methylbutyrate was first detected in the fruit essential oil of *P. anisum* by Kubeczka [23] since this report it has been detected only in *Pimpinella* species [3]. Besides the characteristic phenylpropanoids in *Pimpinella* oils, a number of other C_{12} sesquiterpenes such as geijerene and azulene were also present in considerable amounts [24,25]. From a chemotaxonomic stand point, C_{12} sesquiterpenes and phenylpropanoids are characteristic to the genus *Pimpinella* and are phytochemical markers for this genus that separates them from all the other Apiaceae investigated thus far.

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