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Original Article

Volatile components of *Pimpinella tragium* Vill. from Iran

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Abstract

Volatile components of three parts of *Pimpinella tragium* Vill. (Umbelliferae family) from Iran were investigated qualitatively and quantitatively for the first time.

Essential oils were isolated by hydro-distillation from stems plus leaves, inflorescence and seeds of *P. tragium* individually. The plant materials were collected from Polour (Northeast of Tehran Province).

The yields of the stems plus leaves, inflorescence and seed oils were 0.08%, 0.37% and 1.33%w/w, respectively. Eighteen constituents in the stems plus leaves oil, twenty-six constituents in the inflorescence oil and twenty-three constituents in the seed oil were identified.

Major constituents of the stems plus leaves oil were: germacrene D (34.7%), germacrene B (18.3%), bornyl acetate (15.8%), β-caryophyllene (5.6%) and β-pinene (4.5%). Major constituents of the inflorescence oil were: β-pinene (23.8%), germacrene B (14.1%), hexadecanol (10.3%), β-caryophyllene (7.3%), germacrene D (6.2%) and bornyl acetate (4.1%). Main components of the seed oil were: β-pinene (25.3%), germacrene B (17.8%), sabinene (13.6%), β-caryophyllene (4.8%) and hexadecanol (4.7%).

Keywords: *Pimpinella tragium*; Umbelliferae; Essential oil composition; Germacrene D; Germacrene B; β-pinene.

Introduction

The genus of *Pimpinella* from Umbelliferae family presents 23 species which are found wild in different regions of Iran, two of the more distributed of these species are *P. aurea* and *P. tragium.* The *P. tragium* presents in different regions of Iran, Europe, Anatoli, Turkey, Torkamania, Iraq and north of Africa (1,2).

To the best of our knowledge, there is no previous work concerning the volatile constituents of *P. tragium.* We had reported essential oil composition of *P. aurea* and *P. anisum,* previously (3,4). The yields of stem plus the leaf, inflorescence and seed oils of *P. aurea* were 0.4%, 1.5%, and 2.0%w/w, respectively. Major constituents of the stem plus the leaf oil were found to be 1, 8-cineol, limonene (21.4%), viridiflorol (12.8%), α-pinene (11.5%) and kessane (10.5%). Major constituents of the inflorescence oil were viridiflorol (32.5%) and β-bisabolene (29.5%) while Main constituents of the seed oil were β-bisabolene (50.8%) and viridiflorol (37.0%) (3). Yield of oil seeds of *P. anisum* by steam distillation was 3.3% w/w yield and 11 compound were identified. Main compound was *trans*-anethol (over 90%) (4).

There are some references about essential oil analyses of other *Pimpinella* species in literature (5-13).

The total amount of extractable substances or

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global yield of *P. anisum* seed for the super critical fluid extraction process varied from 3.13-10.67%. The major compound identified and quantified in the extracts was anethol (~90%) (5).

The composition of the essential oil of *Pimpinella serbica* that occurs in Yugoslavia has been reported. The percentage of essential oil ripe fruits was 2.02-3.25%. About 55% of the essential oil was reported to be made up of sesquiterpenes. The main sesquiterpenes were β-caryophyllene (over 47%) and its isomer α humulene (about 2.5%) (6).

Oil Yield of *P. aromatica* aerial parts was 6.1%. Main compounds of aerial parts and fruit oils were methyl chavicol (91.1% and 82.6%) and *trans*-anethol (7.2% and 10%) respectively (7,8).

The oil constituents from the roots, fruits, leaves and stems of *Pimpinella cumbrae* gathered in the Canary Island were investigated. The major constituents in the root oil were found to be isokessane (17%), β-dihydroagarofuran (15%), 2-methyl-butyric acid (10%), geijerene (10%) and pregeijerene (7%). In the fruit oil the main components found were α-bisabolol (39%), δ-3-carene (16%) and limonene (8%). In the leaf oil, α-bisabolol (53%) and δ-3-carene (11%) were the predominant constituents. The most important compounds from the stem oil were α-bisabolol (39%), isokessane (10%) and β-dihydroagarofuran (9%). Pseudoisoeugenol esters were also detected in the oils from the roots, fruits and stems (9).

Yield of stem plus the leaf of *P. squamosa* was 0.17-0.29% and fruit oil yield 4.6-7.0%. Main compounds of whole plant and fruit oils were *trans*-anethol 54.5% and 29.5% respectively (10).

Main compounds of *P. achilleifolia* aerial parts oil (pre-flowering and flowering) were ρ-cymene (52.22%), limonene (9.29%), α-phellandrene (8.76%) and car-2-ene (9.24%) (11).

The aim of this study was to determine the quantity and quality of oils from plant parts of *P. tragium.* For this purpose, oil compositions of three parts of *P. tragium* were characterized.

Experimental

Plant materials

Pimpinella tragium were collected from polour (Northeast of Tehran province, Haraz road, toward Lar, 2300m), at flowering stage in July and seed stage in September 2003 respectively. The fresh plants were dried at room temperature. A herbarium specimen has been deposited in the Herbarium of Research Institute of Forests and Rangelands (TARI).

The dried parts of the plants were crushed to small particle. The average particle size was 0.4 mm. At each time about 80-100 g samples of the stems plus leaves (S&L), inflorescence (IF) and seeds (S) were prepared. The plant material was hydrodistilled for three hours in a Clevenger apparatus to produce the oil. Oil percentages are means of three experiments. The sample oils were dried over sodium sulfate and stored in sealed vials at 4°C temperature before analysis.

GC and GC/MS Analyses

Oils obtained from the (S&L), (IF) and (S) of *P. tragium* were analyzed using a Shimadzu GC-9A gas chromatograph equipped with a DB-5 fused silica column (30 m×0.25 mm, film thickness 0.25 μm, J & W scientific corporation). Oven temperature was held at 60°C for 5 minute and then programmed to 210°C at a rate of 3°C/ min. Injector and detector (FID) temperature were 270°C; helium was used as carrier gas with a linear velocity of 32 cm/s. Percentages were calculated by area normalization method without the use of response factor correction. The retention indices were calculated for all compounds using a homologous series of n-alkanes.

GC/MS analyses were carried out on a Varian 3400 GC/MS system equipped with a DB-5 fused silica column (30 m x 0.25 mm, film thickness 0.25 μm, J & W scientific corporation); oven temperature program was 50°-260°C at a rate of 3°C/min. Transfer line temperature 270°C, carrier gas helium with a linear velocity of 31.5 cm/s, split ratio 1/60, ionization energy 70 ev, scan time 1 sec, mass range 40-300 amu.

Identification of components

The constituents were identified by comparison of their mass spectra with those in a computer library (LIBR-TR and Wiley5 lib.) or with authentic compounds. The identifications were confirmed by comparison of their Retention indices in table I, either with those of authentic compounds or with data in the literature (14).

Results and Discussion

The yields of the (S&L), (IF) and (S) oils were 0.08%, 0.37% and 1.33% w/w, respectively. Therefore the oils yields of generative parts were more than vegetative parts. The oils were pale-yellow in colour.

P. tragium oil yield was campared with other species of *Pimpinella* oils. The oil yields from samples of (S&L), (IF) and (S) of *P. aurea* were respectively 0. 4%, 1.5% and 2.0% w/w. Yield of oil seeds of *P. anisum* and *P. serbica* were respectively 3.13-10.67% and 2-3.2%. Yield of arial part of *P. aromatica* was 6.1% (3-6).

Eighteen constituents in the (S&L) oil, twenty-six constituents in the (IF) oil and twenty-three constituents in (S) oil were identified. Major constituents of the [S&L] oil were: germacrene D (34.7%), germacrene B (18.3%), bornyl acetate (15.8%), β-caryophyllene (5.6%) and β-pinene (4.5%). Major constituents of the [IF] oil were: β-pinene (23.8%), germacrene B (14.1%), hexadecanol (10.3%), β-caryophyllene (7.3%), germacrene D (6.2%) and bornyl acetate (4.1%). Major constituents of the [S] oil were: β-pinene (25.3%), germacrene B (17.8%), sabinene (13.6%), β-caryophyllene (4.8%) and hexadecanol (4.7%).

Other compounds were found in oils but with some quantitative variation that can be seen in Table 1.

Oil composition of *P. tragium* was compared with other species of *Pimpinella*. *Trans*-anetol (90%), β-caryophyllene (47.1%) and β-bisabolene (50.8%) were the major compounds in seed oil of *P. anisum* (4,5), *P. serbica* (6) and

Table1. Percentage of volatile compounds identified in the oil of *Pimpinella tragium.*

Compounds	RI*	Stem+ Leave	inflorescence	Seed
α-pinene	937	0.6	2.5	2.4
sabinene	974	0.5	6.2	13.6
β-pinene	978	4.5	23.8	25.3
6-methyl-5-hepten-2-one	990	-	-	0.4
myrcene	992	0.9	3.0	3.4
α-terpinene	1014	-	0.2	-
ρ-cymene	1015	-	0.2	-
limonene	1029	0.9	1.3	1.1
cis-β-ocimene	1036	-	0.2	-
trans-β-ocimene	1048	-	0.8	0.5
γ-terpinene	1060	0.8	0.4	0.5
trans-pinocarveol	1137	-	0.4	-
terpinen-4-ol	1175	-	0.6	0.5
bornyl acetate	1283	15.8	4.1	1.5
α-yelangene	1370	0.9	-	-
β-caryophyllene	1416	5.6	7.3	4.8
cis-thujopsene	1428	4.0	0.6	-
α-humulene	1452	1.1	1.3	2.6
γ-gurjunene	1471	-	-	1.2
germacrene D	1472	34.7	6.2	-
β-chamigrene	1473	-	2.0	-
γ-muurolene	1474	-	-	4.5
bicyclogermacrene	1492	2.6	1.4	1.2
β-bisabolene	1506	-	-	1.6
δ-cadinene	1522	1.3	1.3	1.1
germacrene B	1554	18.3	14.1	17.8
spathulenol	1574	2.1	1.1	0.5
caryophyllene oxide	1577	1.8	1.1	-
***cis*-3-butyliden-phthalide**	1666	-	1.5	2.6
epi-α-bisabolol	1684	-	-	1.7
hexadecanol	1875	0.7	10.3	4.7
nonadecane	1897	-	2.9	3.2
Total identified		97.1	94.8	96.7

• Retention indices on DB-5 column

Table 2. Type and percentage of different compounds in the oils of *Pimpinella tragium*

Type of compounds	Stem + Leaf		Inflorescence		Seed	
	Number	%	Number	%	Number	%
Monoterpene Hydrocarbone	6	8.2	10	38.6	8	47.2
Oxygenated Monoterpene	1	15.8	3	5.1	2	2.0
Sesquiterpene Hydrocarbon	8	68.5	8	34.2	8	34.8
Oxygenated Sesquiterpen	2	3.9	2	2.2	2	2.2
Aliphatic Hydrocarbon	1	0.7	3	14.7	3	10.5
Total	18	97.1	26	94.8	23	96.7

P. aurea (3) respectively. The major compounds in the aerial parts oil in *P. aromatica* (7), aerial parts and fruit oils in *P. aromatica* (8) were estragol (91%) and methyl chavicol (81.5% and 82.6%) respectively. α-Bisabolol (39%) and ρ-cymene (52.22%) were the major compounds in fruit oil in *P. cumbrae* and aerial parts oil in *P. achilleifolia* (9, 11).

Type and percentage of different compounds in the oils were shown in Table 2. Type of compounds in (IF) and (S) oils were more similar than compounds of (S&L) oil.

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