Essential Oil from Herb and Rhizome of *Peucedanum ostruthium* (L. Koch.) ex DC.

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Peucedanum ostruthium, Essential Oil, Chiral Components

Essential oil from herb and rhizome of *Peucedanum ostruthium* (L.Koch.) ex DC underwent qualitative and quantitative analyses. The content of the oil obtained by hydrodistillation was 0.95% in the herb and 1.25% in the rhizome (per dry weight basis). Gas chromatography (GC) with MS detection and flame ionisation detection showed that the oil from the rhizome contains 39 compounds, of which 29 were identified. Gas chromatography with flame ionisation detection in chiral columns against standard compounds showed the presence of enantiomers of some of the components of the oil. Compounds present in largest quantities are: sabinene (35.2%) of which (+) sabinene accounts for (96.54%) and 4-terpineol (26.6%) of which (+) 4-terpineol accounts for (65.8%). 44 components were found in the herb essential oil, of which 39 compounds were identified. Compounds present in largest quantities were β -caryophyllene (16.1%) and α -humulene (15.8%). The content of sabinene in the herb oil was 4.7%. The following compounds were present in the herb oil only as enantiomers: (+) sabinene (4.7%), (-) limonene (4.4%), (-) β -pinene (0.4%). A coumarin (osthole) was detected in both essential oils (5.5% in herb oil and 5.1% in rhizome oil).

Introduction

Peucedanum ostruthium syn. Imperatoria ostruthium of the Apiaceae family is a perennial plant which grows in the Alps and can also be found in the Sudety mountains (Central Europe, area of Karpacz and Śnieżnik). The rhizome of this plant has been known as a valuable medicinal material, and used in treatment of, among other, conditions of the gall bladder, the liver and the stomach (Madaus, 1938; Muszyński, 1957; Tauscher and Lindequist, 1994).

Earlier studies of the essential oil composition (but not enantiomeric) of this old medicinal plant report the presence of 1.4% essential oil in the rhizome. The oil was reported to contained only d-limonene, dipentene, α -pinene and d-phellandrene (Madaus, 1938; Gildemeister and Hoffmann, 1961). Moreover the other bioconstituents, like flavonoids and coumarines, have been investigated in the above plant material (Hörhammer *et al.*,

1969; Cisowski, 1975; Cisowski et al., 1991; Hiermann et al., 1996; Ganzera et al., 1997).

This paper aims at full quantitative and qualitative analysis of the terpenoids present in the essential oil from the rhizome and also, for comparison, the oil from the herb of *P. ostruthium*. The chromatographic analyses were supposed to be focused on the identification of enantiomeric compounds of the main components of the essential oils.

Materials and Methods

Plant material

The material for the research has been collected in natural sites in the Sudety mountains (Karpacz area). A voucher specimen of *Peucedanum ostruthium* (L. Koch) ex DC (catalogue number 0068) is deposited in the Herbarium of the Medicinal Plant Garden, Medical University of Gdańsk. After drying at room temperature the amount of

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the material was 120 g of the rhizome and 130 g of the herb.

Preparation of essential oils

Dried (room temperature) and pulverised rhizome and herb material (30 g) was hydrodistilled (3h) according to procedures described in Pharmacopea Polonica (FP V, 1995). The essential oils obtained were kept in sealed glass tubes at 4 °C until analysis.

GC-MS (FID) – ITS-40 (Finningan MAT, USA), MS 70EV, 220 °C, GC-FID (Carlo Erba GC 6000, Italy), DD-5 fused silica capillary column (30 m \times 0.25 mm, 0.25 thickness): the temperature programme was 35 °C (2 min.), 4 °C/min., 300 °C (15 min.); helium flow rate 1 ml/min.

GC-FID – Hewlett-Packard 5890 series II,chiral column-permethyl- β -cyclodextrin-. 120 (Supelco) 30 m × 0.25 mm ID, the temperature: 70 °C isotherm, argon 100 kPa.

Results and Discussion

Comparative GC-MS analysis of *Peucedanum* ostruthium herb and rhizome was carried out as far as essential oil composition was concerned. In the course of phytochemical analyses DD-5 fused silica capilary column (achiral) and a chiral one (permethyl- β -cyclodextrin) were used. This procedure made it possible to carry out chromatographic analyses of the full set of terpenoid compounds present in the essential oils of the researched plant materials.

The enantiomeric separation of chiral terpenoids in *Peucedanum ostruthium* essential oil seems to be of a great importance. It is well known that stereospecifity of drug action is related to the molecular asymmetry of the component receptors of the human body. As a consequence, the diastereomers and enantiomers present in the natural mixtures of volatile oils have significantly different biological activities (Sybilska *et al.*, 1994).

To achieve the desired separation of the enantiomeric compounds the capilary column with the permethyl- β -cyclodextrin stationary phase was used. This chromatographic system has been successfully used for the separation of monoterpenes in the essential oils of several species (Sybilska *et al.*, 1994).

The essential oils from rhizome and herb were similar in terms of qualitative composition, although some differences were noted. Table I gives the content of the terpenoid compounds (calculated as % of the total oil) present in the investi-

Table I. Percentage content of compounds in essential oils in the herb and rhizome of *Peucedanum ostruthium* (L.Koch.) ex DC.

Compound	RI	Percentage content Herb Rhizome	
α-Thujene	924.0	t	0.7
α-Pinene	929.4	3.1	0.7
Camphene	944.0	0.5	t
Sabinene	970.6	4.7	35.2
β-Pinene	972.8	0.4	1.0
Myrcene	979.8	3.1	1.3
α-Phellandrene	1001.9	0.2	3.7
α-Terpinene	1013.1	t	1.9
p-Cymene	1023.3	0.4	0.2
Limonene+ β -Phellandrene	1026.5	4.4	2.8
cis-Ocimene	1037.6	6.4	t
Trans-Ocimene	1047.9	0.7	t
γ-Terpinene	1057.7	0.1	3.0
Trans-Sabinene hydrate	1068.6	t	0.8
Terpinolene	1087.2	t	0.9
cis-Sabinene hydrate	1099.8	t	0.8
cis-p-menth-2-n-1-ol	1123.1	0.1	1.9
Trans-cis-p-menth-2-n-1-ol	1142.1	t	0.7
Trans-Verbenol	1147.5	0.3	_
4-Terpineol	1178.7	1.5	26.6
α-Terpineol	1193.6	t	0.6
≥-Elemene	1337.7	0.3	1.1
α-Ylangene	1372.4	0.4	0.2
α-Copaene	1376.8	0.5	t
β-Bourbonene	1386.2	0.7	_
β-Cubebene	1391.4	0.3	_
β-Elemene	1392.9	1.1	0.8
β-Caryophyllene	1423.2	16.1	0.1
α-Humulene	1458.2	15.8	t
β -Cadinene	1479.9	0.2	t
Germacrene D	1485.4	9.6	0.8
unknown sesquiterpene	1495.0	3.5	_
Germacrene B	1500.5	2.2	3.0
α-Fanesene	1508.5	3.9	_
γ-Cadinene	1518.1	0.5	0.4
>-Cadinene	1526.9	1.2	0.7
Kessane	1526.9	t	0.9
4-β-hydroxygermacra-1(10),5- diene	1581.6	0.6	t
Spathulenol	1584.9	1.4	t
Caryophyllene oxide	1590.1	1.6	t
Unknown sequiterpene $M = 220$		1.0	_
Germacrene D-4-ol	1627.8	0.3	0.3
γ-Cadinol	1648.1	0.5	0.1
α-Cadinol	1622.3	1.0	t
Osthole	2156.0	5.5	5.1

t, traces.

RI, retention index

gated plant materials, determined by using an achiral column. The following 6 components were present in the herb oil: transverbenol (0.3%), β bourbonene (0.7%), β -cubebene (0.3%), α -farnesene (3.9%), and an unknown sesquiterpene (1.0%), none of which was present in the P.ostruthium rhizome oil. There are very clear differences in the quantitative composition of the oils (Table I). The main components of the herb oil are: β -caryophyllene (16.1%) and α -humulene (15.8%), which are only present in small quantities in the rhizome oil (0.1% and traces respectively). The main components in the rhizome oil are: sabinene (35.2%) and 4-terpineol (26.6%) which are present in the herb oil in small quantities only (sabinene 4.7% and 4-terpineol 1.5%).

The enantiomeric composition of the terpene fraction in the tested plant material is demonstrated in Table II. As far as enantiomeric forms are concerned, sabinene was present in both oils as (+) sabinene (96% in rhizome oil and 100% in herb oil). There was also a number of other hydrocarbons present in the herb essential oil only as enantiomeric forms. These were: (-) limonene (4.4%) and (-) β -pinene (0.4%). In addition, there was a high amount of (-) α -pinene (72.4% of 3.1% α -pinene) and (+) camphene (69% of 0.5% camphene) (Table II).

Apart from (+) sabinene and (+) terpinen-4-ol the rhizome oil contained high amounts of the following enantiomeric compounds: (-) limonene

Table II. Content of enantiomeric forms per 100% of the compound content in the oil of . *Peucedanum ostruthium* (L. Koch.) ex DC.

Compound	Rt	Percentage content Herb Rhizome	
(-)-α-Pinene	21.32	72.4	41.5
(+)-α-Pinene	22.16	27.6	58.5
(-)-Sabinene	28.45	_	3.5
(+)-Sabinene	24.73	100	96.5
(-)-Limonene	37.07	100	89.4
(+)-Limonene	40.35	_	10.6
(-)-4-Terpineol	40.32		34.1
(+)-4-Terpineol	39.28		65.9
(-)-Camphene	25.73	31.0	_
(+)-Camphene	26.31	69.0	-

Rt, retention time.

(88.7% of 2.8% limonene) and (+) α -pinene (59.1% of 0.7% pinene).

In the rhizome essential oil, unlike the herb oil, no pure enantiomers were identified against standard compounds, but only the mixtures. Since pure enantiomeric forms are present in the *P.ostruthium* herb oil, it can serve as a set of standard substances for these particular forms to be used for comparative purposes in chromatography. Moreover the results of the thorough chromatographic analysis (chiral columns, large library of standard compounds) can be used , in future, for the interpretation of the pharmacological activity of the plant.

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