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Chemical composition of *Echinophora tenuifolia* subsp. *sibthorpiana* essential oil from Greece

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Abstract: Aerial parts of *Echinophora tenuifolia* subsp. *sibthorpiana* were subjected to hydrodistillation and the oil obtained was analyzed by GC and GC-MS. The essential oil was dominated by the presence of monoterpenes with α-phellandrene (43.8%) being the major component, followed by the high percentage of the phenylpropanoid derivative methyleugenol (28.6%), while sesquiterpenes were not detected.

Keywords: *Echinophora tenuifolia*; essential oil composition; α-phellandrene; methyleugenol.

1. Introduction

The genus *Echinophora* (fam. Umbelliferae) comprises about 10 species, ranging from the Mediterranean region eastwards to Afghanistan [1]. There are three taxa of the genus distributed in Europe (*E. spinosa* L., *E. tenuifolia* L. subsp. *sibthorpiana* (Guss.) Tutin and *E. tenuifolia* L. subsp. *tenuifolia*) [2]. *E. tenuifolia* subsp. *sibthorpiana* is a greyish-pubescent perennial herb, to 20-50 cm tall, with yellow petals. It grows in dry places and is native to Greece and Kriti to Krymea [2]. According to Dioscorides the seeds and the root of *E. tenuifolia* are efficient in the treatment of epilepsy [3]. In continuation of our research on the essential oils of Greek plants, we have investigated the volatile constituents of the aerial parts of *E. tenuifolia* subsp. *sibthorpiana.*

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2. Materials and Methods

2.1. Plant Material

Aerial parts of *E. tenuifolia* subsp. *sibthorpiana* were collected during the flowering stage in October, 2007 from Sourpi (Magnesia Prefecture), Greece. The plant sample was identified by Dr Bazos and a voucher specimen (OT-80) was deposited in the Herbarium of the University of Athens (ATHU). Fresh plant material was chopped and subjected to hydrodistillation for 3 hours using a modified Clevenger apparatus to obtain the essential oil, and subsequently it was dried over anhydrous sodium sulphate and, after filtration, stored under N_2 atmosphere in amber vials at 4 ^{o}C until analysis. The oil yield was 1.53% (w/v)

2.2 Gas Chromatography

GC analysis was carried out using a SRI 8610C GC-FID system, equipped with DB-5 capillary column (30 m x 0.32 mm; film thickness 0.25 μm) and connected to a FID detector. The injector and detector temperature was 280 ^{o}C. The carrier gas was He, at flow rate of 1.2 mL/min. The thermal program was 60 ^{o}- 280 ^{o}C at a rate of 3 ^{o}C/min; split ratio 1:10. Two replicates of the oil were processed in the same way.

Analysis of the essential oil was performed using a Hewlett Packard 5973-6890 GC-MS system operating in the EI mode at 70eV, equipped with a split/splitless injector (200 ^{o}C). The transfer line temperature was 250 ^{o}C. Helium was used as carrier gas (1 mL/min) and the capillary column used was HP 5MS (30 m x 0.25 mm; film thickness 0.25 μm). The temperature program was the same with that used for the GC analysis; split ratio 1:10. The injected volume was 1μL. Total scan time 83.33 min. Acquisition mass range 40-400 amu. The identification of the compounds was based on comparison of their retention indices (RI), their retention times (RT) and mass spectra with those obtained from authentic samples (purchased from the Sigma-Aldrich Co) and/or the NIST/NBS, Wiley libraries and the literature [4].

3. Results and Discussion

The chemical composition of *E. tenuifolia* subsp. *sibthorpiana* essential oil is summarized in Table 1. Twenty-eight compounds were characterized representing 98.6% of the total oil. The oil was characterized by the occurence of monoterpenes with *α*-phellandrene (43.8%) being the dominant component. The phenylpropanoid derivative methyleugenol was also presented in the oil with a high percentage (28.6%). The absence of sesquiterpenes in the oil was notable.

The essential oil composition of *E. tenuifolia* subsp. *sibthorpiana* of Greek origin is quite similar to the composition of other *E. tenuifolia* subsp. *sibthorpiana* samples collected from Turkey and Iran. According to previous reports on the analysis of aerial parts volatile constituents of *E. tenuifolia* subsp. *sibthorpiana*, the phenylpropanoid derivative methyleugenol (amounting to 80.6%) and the monoterpenes *α*-phellandrene (amounting to 51.5%), and δ-3-carene (amounting to 36.6%) were the most abundant [5-9]. α-Phellandrene and methyleugenol were reported by most of the authors as the main constituents of *E. tenuifolia* subsp. *sibthorpiana* oil. It is noteworthy that Chalchat et al. (2007) did not indicate α-phellandrene, but δ-3-carene was among the main constituents of samples oils of Turkish origin. Furthermore in previous studies the monoterpenes β-phellandrene and p-

cymene were reported as main metabolites in the oils [5-7]. The latter components were also present in the oil of Greek origin in considerable amounts (7.4% and 9.5%, respectively). Akgul and Chialva (1989) have also reported camphene as an important constituent (27.1%), which was not detected in Greek *E. tenuifolia* subsp. *sibthorpiana* oil. It is probable that the geographic area of the plant collection and the vegetation stage of the plant are among the factors that effect on the oil yield and the composition of this subspecies.

Table 1. Chemical composition of the essential oil of *Echinophora tenuifolia* subsp. *sibthorpiana*

Compound	**RI**	**Composition (%)**
n-nonane	900	t
α-thujene	925	1.0
α-pinene	931	3.1
sabinene	970	0.9
β-pinene	975	t
myrcene	986	t
α-phellandrene	997	43.8
α-terpinene	1012	0.1
p-cymene	1020	9.5
β-phellandrene	1025	7.4
(*Z*)-β-ocimene	1032	t
γ-terpinene	1055	2.3
terpinolene	1084	0.9
6,7-epoxymyrcene	1088	0.4
dehydro-sabinaketone	1116	0.2
(*Z*)-epoxy-ocimene	1130	t
(*E*)-epoxy-ocimene	1140	t
(2*Z*)-nonen-1-al	1143	t
isoborneol	1157	0.1
borneol	1164	0.3
terpinen-4-ol	1172	t
cryptone	1180	t
dill ether	1181	t
α-phellandrene epoxide	1192	0.4
octanyl acetate	1204	t
methyleugenol	1394	28.6
myristicin	1509	t
elemicin	1547	t
Total		99.0
Grouped components		
Monoterpene hydrocarbons		69.0
Oxygenated monoterpenes		1.4
Phenylpropanoids		28.6

[a]RI= retention indices relative to C_9-C_{23} *n*-alkanes on the HP-5MS column; t= trace (<0.1%)

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