

**KANADA'DA YETİŞEN İKİ *Thuja* L. (CUPRESSACEAE)
TÜRÜNÜN UÇUCU YAĞ KOMPOZİSYONU**

**ESSENTIAL OIL COMPOSITION OF TWO *Thuja* L.
(CUPRESSACEAE) SPECIES FROM CANADA**

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ÖZET

Kanada'da yetişen *Thuja occidentalis* L. ve *Tetraclinis articulata* (Vahl) Masters (synonym: *Thuja articulata*) türlerinin ibrelerinden elde edilen yağlar HS-SPME / GC-MS tekniği ile analiz edildi. Sırasıyla %92.30 and %93.42'lik toplam yağ miktarında otuzbir ve ellibir bileşen tespit edildi. *Thuja occidentalis*'in ana bileşenleri bornilasetat (%30.00), limonen (%7.56), kamfor (%7.33), α -pinen (%7.18) ve δ -kadinen (%6.01) olarak belirlendi; *Tetraclinis articulata*'da ise α -pinen (%32.67), 3-karen (%18.29), β -mirsen (%11.69) ve bornilasetat (%5.88) ana bileşenler olarak bulundu. Sonuçlara göre *T. occidentalis*'in kemotipi bornilasetat, *T. articulata*'nın kemotipi α -pinen olarak belirlenmiş ve HS-SPME yöntemi tıbbi ve aromatik bitkilerin rutin kontrol ve analizlerinde kullanılabilir bir yöntem olarak belirlenmiştir.

Anahtar kelimeler: *Thuja occidentalis*; *Tetraclinis articulata*; uçucu yağ; HS-SPME

ABSTRACT

The essential oil needles of *Thuja occidentalis* L. and *Tetraclinis articulata* (Vahl) Masters (synonym: *Thuja articulata*) growing in Canada, were analyzed by HS-SPME / GC-MS technique. Thirty one and fifty one compounds were identified representing 92.30% and 93.42% of the oil, respectively. The main constituents of *Thuja occidentalis* were bornylacetate (30.00%), limonene (7.56%), camphor (7.33%), α -pinene (7.18%) and δ -cadinene (6.01%), whereas α -pinene (32.67%), 3-carene (18.29%), β -myrcene (11.69%) and bornylacetate (5.88%) were the major constituents of *Tetraclinis articulata*. The results showed that *T. occidentalis* chemotype was bornylacetate whereas α -pinene was chemotype of *Tetraclinis articulata* and HS-SPME method can be applied to routine control analysis of aromatic and medicinal plants.

Key Words: *Thuja occidentalis*; *Tetraclinis articulata*; essential oil; HS-SPME

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INTRODUCTION

T. occidentalis L. (Cupressaceae), commonly known as Arbor vitae or white cedar, is indigenous to eastern North America and is grown in Europe as an ornamental tree [1]. The plant was first identified as a remedy by native Indians in Canada during a 16th century expedition and was found to prove effective in the treatment of weakness from scurvy [2]. *T. occidentalis* is an evergreen species widely cultivated as a common ornamental plant [3]. During the last years most of the phytochemical studies on this plant species focused on the terpene composition of the essential oil, where α -pinene and α -cedrol have been reported as the major constituents of the leaf oil [4,5]. In folk medicine, *T. occidentalis* has been used to treat bronchial catarrh, enuresis, cystitis, psoriasis, uterine carcinomas, amenorrhea and rheumatism [6,7].

T. occidentalis was originally cultivated in North America. It is a native European tree with a maximal height of 15-20 m. It has coniferous pyramidal features, with flattened branches and twigs in one plane, bearing small scale-like leaves. Over the whole year, the leaves are green, with the lower side showing a brighter green where resin glands also reside. Small, 1-2 cm long green to brown coniferous pins contain the seeds [8]. In addition, some data about biology, pharmacology, toxicology, odor and use in food-flavorings, perfumery and cosmetics as well as medical applications of main compounds (α - and β -thujone, camphor, 1,8-cineole, linalool and linalyl acetate) were published elsewhere [9,10]. The aim of the present study is to provide chemical data that might be helpful in potential usefulness, to summarize the available information in order to facilitate and guide future researches and to examine potential chemotaxonomic significance of these species.

MATERIAL AND METHODS

Plant Source

T. occidentalis was collected in vicinity of Toronto / Canada, 27.05.2012, 100-200 m., Kilic 4454. *T. articulata* was collected in vicinity of Toronto / Canada, 27.05.2012, 100-200 m.,

Kilic 4455.

HS-SPME Procedure

Five grams powder of needles of *T. occidentalis* and *T. articulata* were carried out by a (HS-SPME) head space solid phase microextraction method using a divinyl benzene/ carboxen / polydimethylsiloxane (DVB/CAR/PDMS) fiber, with 50/30 μ m film thickness; before the analysis the fiber was pre conditioned in the injection port of the gas chromatography (GC) as indicated by the manufacturer. For each sample, 5 g of plant samples, previously homogenized, were weighed in to a 40 ml vial; the vial was equipped with a "mininert" valve. The vial was kept at 35°C with continuous internal stirring and the sample was left to equilibrate for 30 min; then, the SPME fiber was exposed for 40 min to the headspace while maintaining the sample at 35°C. After sampling, the SPME fiber was introduced into the GC injector, and was left for 3 min to allow the analytes thermal desorption. In order to optimize the technique, the effects of various parameters, such as sample volume, sample headspace volume, sample heating temperature and extraction time were studied on the extraction efficiency as previously reported by Verzera *et al.*, [11].

GC-MS Analysis

A Varian 3800 gas chromatograph directly interfaced with a Varian 2000 ion trap mass spectrometer (VarianSpa, Milan, Italy) was used with injector temperature, 260°C; injection mode, splitless; column, 60 m, CP-Wax 52 CB 0.25 mm i.d., 0.25 μ m film thickness (ChrompackItalys.r.l., Milan, Italy). The oven temperature was programmed as follows: 45°C held for 5 min, then increased to 80°C at a rate of 10°C/min, and to 240°C at 2°C/min. The carrier gas was helium, used at a constant pressure of 10 psi; the transfer line temperature, 250°C; the ionisation mode, electron impact (EI); acquisition range, 40 to 200 m/z; scan rate, 1 us⁻¹. The compounds were identified using the NIST (National Institute of Standards and Technology) library (NIST/WILEY/EPA/NIH), mass spect-

ral library and verified by the retention indices which were calculated as described by Van den Dool and Kratz [12]. The relative amounts were calculated on the basis of peak-area ratios. The chromatograms of studied samples are seen in Figure 1,2 and identified constituents are listed in Table 1.

Table 1. Chemical composition of the studied samples

Constituents	RRI	<i>Thuja occi-</i> <i>dentalis</i>	<i>Thuja arti-</i> <i>culata</i>
α -pinene	1022	7.18	32.67
Camphene	1049	3.05	1.71
β -pinene	1068	3.60	-
3-carene	1076	-	18.29
β -myrcene	1090	-	11.69
Limonene	1107	7.56	5.58
α -terpinolene	1153	-	1.57
<i>p</i> -cymene	1180	-	0.48
2,4,6-octatriene	1244	-	0.07
1,3,5-undecatriene	1257	-	0.80
<i>p</i> -dimethylstyrene	1304	-	0.26
Benzene, 1-methyl-4	1312	-	0.59
α -cedrene	1321	-	0.79
1,3-cyclohexadiene	1332	-	0.20
Sabinenehydrate	1351	-	0.20
7-methanoazulene	1370	-	0.15
Limoneneoxide	1383	2.08	2.41
1,6-octadien-3-ol	1386	0.65	-
β -funebrene	1390	0.48	0.79
β -elemene	1395	1.59	-
β -cedrene	1399	-	0.11
Thujone	1404	0.65	-
β -sesquiphellandrene	1408	0.46	-
γ -elemene	1409	-	0.55
Thujopsene	1428	-	0.11
Caryophyllene	1433	0.56	0.34
3-cyclopentene-1-ace- taldehyde	1436	-	0.20
β -farnesene	1442	0.21	0.62
1,3-cyclohexadiene	1454	0.26	-
Cyclopentane	1455	-	0.31
γ -cadinene	1458	0.96	-
α -cedrene	1460	-	1.14
Bornylacetate	1477	30.00	5.88
Benzene, 1-methyl-4	1481	-	0.20
β -bisabolene	1483	-	0.27

Isobornylacetate	1489	-	0.21
Exo-methyl-camp- henilol	1498	-	0.08
Camphor	1508	7.33	1.05
δ -cadinene	1510	6.01	-
β -selinene	1519	-	0.18
α -amorphene	1520	4.43	-
Ethanol	1527	0.66	0.09
Verbenol	1535	1.45	-
β -sesquiphellandrene	1551	-	0.09
α -curcumene	1561	0.64	-
Benzene	1562	-	0.79
Borneol	1577	5.56	0.81
Myrtenol	1588	1.11	0.05
1,4-cyclohexadiene	1598	1.01	0.12
Benzene, 1-methyl-4	1652	-	0.23
Carveol	1680	0.26	0.10
Quinoline	1688	-	0.04
Adamantane	1729	0.21	0.20
Benzenemethanol	1736	-	0.21
Verbenone	1741	0.78	0.18
γ -curcumene	1811	0.40	-
1,6,10-dodecatrie- ne-3-ol	1823	-	0.04
Caryophylleneoxide	1936	0.19	0.13
di-epi- α -cedrene	1958	-	0.16
Cedrol	1962	1.06	0.37
Bicyclo(2.2.1)hepta- ne2,5-dione	2048	-	0.10
Phenanthrene	2182	-	0.01
1,3-benzenediamine	2565	1.91	0.20
	Top- lam	92.30	93.42

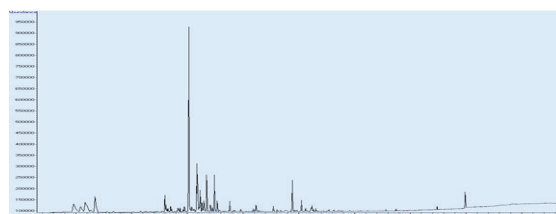


Figure 1. GC-MS chromatogram of *T. occi-*
dentalis from Canada extracted by HS-SPME.

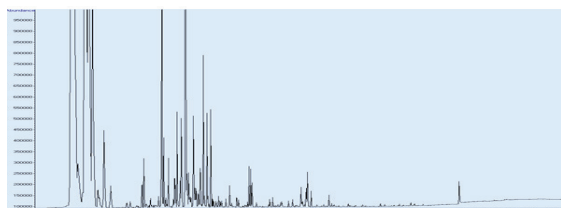


Figure 2. GC-MS chromatogram of *T. articulata* from Canada extracted by HS-SPME.

RESULTS AND DISCUSSION

The essential oils of 31 compounds were identified representing 92.30% of the oils; bornylacetate was determined to be present at the high percentage (30.00%), the presence of limonene (7.56%), α -pinene (27.72%), camphor (7.33%) and δ -cadinene (6.01%) were also important for the oil profile (Table 1). The major components of the essential oil from *T. occidentalis* cultivated in Iran were α -pinene (21.9%), α -cedrol (20.3%), Δ -3-carene (10.5%) and limonene (7.2%) [13].

Volatile constituents *Thuja articulata*, 51 components were identified representing 93.42% of the oil, α -pinene was the predominant compound (32.67%) followed by 3-carene (18.29%), myrcene (11.69%) and bornylacetate (5.88%) (Table 1). α -pinene (29.2%), Δ -3-carene (20.1%), α -cedrol (9.8%), caryophyllene (7.5%), α -humulene (5.6%), limonene (5.4%), α -terpinolene (3.8%) and α -terpinyl acetate (3.5%) were the main constituents in the essential oil of *T. orientalis* [14].

In another study with six *Pinus* L. taxa; the main compounds of studied taxa were: *P. resinosa*; caryophyllene (27.60%), α -pinene (12.96%), 3-carene (12.93%) and naphthalene (9.37%). *P. flexilis*; α -pinene (33.29%), β -pinene (16.24%) and germacrene D (6.13%). *P. nigra*; acetic acid (31.12%), bicyclo [2.2.1] heptan-2-one (21.45%) and borneol (8.64%). *P. strobus*; α -pinene (32.96%), β -myrcene (27.72%) and β -pinene (8.01%). *P. parviflora*; α -pinene (25.56%), caryophyllene (13.21%),

germacrene D (6.71%), limonene (6.21%) and camphene (5.68%). *P. mugo* subsp. *mugo*; 3-carene (36.54%), *p*-cymene (18.03%), α -pinene (9.00%) and limonene (5.09%) [15]. *Juniperus communis* L., *Taxus canadensis* Marshall. and *Tsuga canadensis* (L.) Carr. from Canada were investigated by microextraction HS-SPME / GC-MS and limonene (26.12%), benzene (15.62%), β -myrcene (9.08%) and β -pinene (7.30%) were detected the main constituents of *J. communis*; 1-propanone (36.38%), morpholine (10.95%), methylamine (9.10%) and methanone (8.14%) were found to be main components of *Taxus canadensis*; bornylacetate (26.84%), α -pinene (23.74%), camphene (11.93%) and limonene (6.02%) were determined the major constituents of *Tsuga canadensis* [16]. On the other hand, in this study the main constituents of *Thuja occidentalis* were bornylacetate (30.00%), limonene (7.56%), camphor (7.33%), α -pinene (7.18%) and δ -cadinene (6.01%); α -pinene (32.67%), 3-carene (18.29%), β -myrcene (11.69%) and bornylacetate (5.88%) were found to be the major constituents of *Tetraclinis articulata*.

According to these results, it is possible to say that, *Thuja occidentalis* and *Thuja articulata* both are showed α -pinene chemotype essential oils.

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