Essential Oil Composition of *Laurus nobilis* L. of Different Growth Stages Growing in Iran

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The chemical variations of the essential oil from the aerial parts of *Laurus nobilis* L. (Lauraceae) have been studied. Plant material has been harvested at each phenological status (vegetative, before anthesis, full flowering and seed-bearing). The oils were obtained by hydrodistillation of the air-dried samples. Analysis by GC and GC-MS of the essential oils has allowed to identify 39 compounds. The main compounds were 1,8-cineole, *trans*-sabinene hydrate, α-terpinyl acetate, methyl eugenol, sabinene, eugenol and α-pinene.

Key words: Laurus nobilis L., Lauraceae, 1,8-Cineole

Introduction

Laurus nobilis L. (Lauraceae) is an evergreen shrub indigenous to the south parts of Europe and the Mediterranean area. It is cultivated in the north of Iran (Zargari, 1990). In Iranian folk medicine, the leaves of this plant have been used to treat epilepsy (Aqili-khorasani, 1992; Zargari, 1990), neuralgia and parkinsonism (Aqili-khorasani, 1992). The essential oil obtained from its leaves has been used for relieving hemorrhoid and rheumatic pains (Zargari, 1990). It also has diuretic (Aqili-khorasani, 1992; Zargari, 1990), antifungal (Qamar and Chaudhary, 1991) and antibacterial (Seyed *et al.,* 1991) activities.

There are many studies on the chemical composition of the essential oil obtained from leaves of Mediterranean and European *L. nobilis* (Riaz *et al.,* 1989; Lin *et al.,* 1990; Baghdadi *et al.,* 1993; Putievsky *et al.,* 1994; Fiorini *et al.,* 1997). In the study of Riaz *et al.* (1989), the main components of the essential oil were cineole (44.1 %), eugenol (15.2 %), sabinene (6.2 %), 4-terpineol (3.6 %), α-pinene (2.7 %), methyl eugenol (2.5 %), α-terpineol (2.2 %), and β-pinene (2.1 %). Pharmacological studies have demonstrated the anesthetic, hypothermic, muscle-relaxant and anticonvulsant activities of eugenol and methyl eugenol (Dallmeier and Carlini, 1981) and also the antistress effect of eugenol (Sen *et al.,* 1992). Furthermore, some analogues of α-pinene prevent the audiogenic seizures in susceptible rats (Consroe *et al.,* 1981).

To the best of our knowledge, an investigation of the essential oil of *Laurus nobilis* L. from Iran has not been reported to date. As a part of our studies on the chemical composition of essential oils and the screening program for bioactive compounds from plants that grow in Iran, the present study describes the essential oil composition of aerial parts of *Laurus nobilis* L. in different growth stages.

Materials and Methods

Plant material and chemicals

The aerial parts of *Laurus nobilis* L. were collected at four different stages of development, respectively, in May (vegetative plants), August (before anthesis), September (full flowering plants) and November (seed-bearing plants) of 2006 at Tabriz, northwest of Iran. Voucher specimens were deposited at the Herbarium of Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran.

All solvents applied were of pro-analysis purity and were purchased from Fluka Chemical Co. (Buchs, Switzerland). Anhydrous sodium sulfate was obtained from Merck (Darmstadt, Germany).

Isolation of the essential oil

The aerial parts (100 g) were dried at 25 °C in the shade and subjected to hydrodistillation using a Clevenger-type apparatus for 4 h. The oil was dried with anhydrous sodium sulfate, weighed and stored at 4–6 °C in dark until use.

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Gas chromatography-mass spectrometry (GC-MS)

A Hewlett-Packard 6890 gas chromatograph with a HP-5 capillary column (phenyl methyl siloxane of 25 m length, 0.25 mm i. d., and 0.25 μm film thickness) was used. Carrier gas was He; split ratio was 1:25, and the detection was done by flame ionization. The temperature program was: 60 °C for 2 min, rising to 240 °C at 4 °C/min. Injector temperature was 250 °C, and detector temperature was 260 °C. GC-MS was done by a Hewlett-Packard 6859 instrument, equipped with a quadrup detector, on a HP-5 column (see GC), operating at 70 eV ionization energy, using the same temperature program and carrier gas as above. Retention indices were calculated by comparing the retention times with those of C_8–C_{22} samples that were injected after the oil at the same chromatographic conditions according to the method of Van Den Dool and Kratz (1963).

Identification of compounds

The linear retention indices for all compounds were determined by coinjection of the sample with

Table I. Chemical composition of *Laurus nobilis* L. essential oil.

Compound	RI	Phenological stage[a]				Standard deviation
		Vegetative	Buds	Flowers	Seeds	
α-Thujene	933	0.4	1.0	0.3	0.3	0.33
α-Pinene	941	3.2	2.6	2.6	2.7	0.28
Camphene	955	0.3	0.3	0.3	0.3	0
Sabinene	977	6.5	6.0	5.8	5.9	0.31
β-Pinene	981	2.9	2.5	2.4	2.5	0.22
Myrcene	992	1.1	0.9	0.8	1.0	0.12
3-Carene	1013	0.5	0.4	0.4	0.5	0.05
α-Terpinene	1020	0.2	0.2	0.2	0.2	0
p-Cymene	1028	0.2	0.2	0.2	0.2	0
Limonene	1032	1.3	1.4	1.3	1.3	0.05
1,8-Cineole	1035	35.7	34.9	31.4	35.7	2.05
γ-Terpinene	1064	0.4	0.4	0.3	0.4	0.05
cis-Sabinene hydrate	1070	0.6	0.6	0.6	0.6	0
Terpinolene	1089	0.2	0.2	0.2	0.2	0
trans-Sabinene hydrate	1100	9.7	11.9	9.8	11.4	1.11
δ-Terpineol	1166	–	0.4	0.6	0.4	0.25
Borneol	1167	–	0.2	–	0.2	0.11
4-Terpineol	1180	1.4	1.6	1.6	1.5	0.09
α-Terpineol	1190	2.8	3.2	3.3	3.0	0.22
Nerol	1227	0.2	0.3	0.3	0.2	0.05
Linalyl acetate	1258	0.3	0.4	0.4	0.4	0.05
Isobornyl acetate	1286	0.3	0.4	0.4	0.4	0.05
Terpinen-4-yl acetate	1291	0.1	0.2	0.2	0.2	0.05
α-Terpinyl acetate	1351	9.3	12.1	11.4	10.4	1.21
Eugenol	1356	4.8	3.8	5.5	4.3	0.72
β-Elemene	1390	0.1	–	–	–	0.05
Methyl eugenol	1404	6.8	8.1	9.4	7.9	1.06
β-Caryophyllene	1420	0.6	0.4	0.5	0.5	0.08
Valencene	1492	0.3	0.3	0.4	0.3	0.05
β-Bisabolene	1509	0.2	–	0.2	0.2	0.1
Elemicin	1554	0.6	0.5	0.7	0.5	0.09
Spathulenol	1577	0.6	–	–	–	0.3
Germacrene D	1574	1.2	1.2	1.6	1.2	0.2
Caryophyllene oxide	1581	0.2	0.7	0.8	0.7	0.27
Viridiflorol	1590	0.3	0.2	0.3	0.2	0.05
γ-Eudesmol	1630	0.2	0.1	0.2	0.2	0.05
Bisabolol	1637	0.2	0.2	0.2	0.2	0
β-Eudesmol	1647	0.5	0.5	0.5	0.5	0
α-Cadinol	1652	1.2	0.8	1.3	1.0	0.22
Total identified		95.8	98.8	95.5	97.3	

[a] At each stage, three samples have been analyzed.

Table II. Percentages of the main chemical classes of volatiles.

Chemical class	Phenological stage			
	Vegetative	Buds	Flowers	Seeds
Oxygenated monoterpenes	60.43	66.19	59.87	64.32
Monoterpene hydrocarbons	17.37	16.11	14.82	15.35
Phenylpropanoids	1.24	12.40	15.58	12.67
Sesquiterpene hydrocarbons	1.22	0.66	1.06	1.04
Oxygenated sesquiterpenes	4.76	3.60	4.38	3.94

a solution containing the homologous series of C_8–C_{22} *n*-alkanes. The individual constituents were identified by their retention indices, referring to known compounds from the literature (Adams, 1995) and also by comparing their mass spectra with either those of the known compounds or with those of the Wiley mass spectral database.

Results and Discussion

The essential oil contents of the aerial parts of *Laurus nobilis* L., obtained by hydrodistillation, were 0.7 %, 0.8 %, 1.1 % and 0.6 % in the vegetative, bud, flowering and seed-bearing stages, respectively, calculated on a dry weight basis. The components of the essential oils are reported in Table I. Thirty-seven components accounting for 95.8 % of the total composition were identified in the vegetative stage. The major constituents of this oil were 1,8-cineole (35.7 %), *trans*-sabinene hydrate (9.7 %), α-terpinyl acetate (9.3 %), methyl eugenol (6.8 %), sabinene (6.5 %) and eugenol (4.8 %). In the volatile of the bud stage, thirty-six compounds amounting 98.8 % of the total components were identified which included 1,8-cineole (34.9 %), α-terpinyl acetate (12.1 %), *trans*-sabinene hydrate (11.9 %), methyl eugenol (8.1 %), sabinene (6.0 %) and eugenol (3.8 %) as main components. In the oil obtained from the flowering stage, thirty-six components were identified, which represented about 95.5 % of the total composition. 1,8-Cineole (31.4 %), α-terpinyl acetate (11.4 %), *trans*-sabinene hydrate (9.8 %), methyl eugenol (9.4 %), sabinene (5.8 %) and eugenol (5.5 %) were the principal components of this oil. In the seed-bearing stage, thirty-seven constituents accounting for 97.3 % of the total oil were identified that included 1,8-cineole (35.7 %), *trans*-sabinene hydrate (11.4 %), α-terpinyl acetate (10.4 %), methyl eugenol (7.9 %), sabinene (5.9 %) and eugenol (4.3 %) as main components. The majority of the identified compounds belonged to the monoterpene fraction (Table II), with percentages ranging from 77.8 % in the vegetative stage, to 82.3 % before anthesis, 74.7 % in the flowering stage and 79.7 % in the seed-bearing stage. The oxygenated fraction was mainly composed of monoterpenes and the bud stage oil had the highest percentage of oxygenated monoterpenes.

The results from this study show that oils obtained from the different phenological stages have nearly similar compositions; the main compounds were 1,8-cineole, *trans*-sabinene hydrate, α-terpinyl acetate, methyl eugenol, sabinene, eugenol, α-pinene, and α-terpineol. Thus the time of harvesting of this plant does not have a major effect on the chemical composition of the essential oil, but it affects the essential oil content of the plant. The flowering stage is the best time for harvesting the plant because at this time the plant contains the highest percentage of essential oil.

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