Chemistry of Natural Compounds, Vol. 42, No. 6, 2006

CHEMICAL COMPOSITION AND ANTIMICROBIAL ACTIVITY OF THE ESSENTIAL OILS OF *Ferula latisecta* AND *Mozaffariania insignis* FROM IRAN

Zohreh Habibi,[1*] Peyman Salehi,[2] Maryam Yousefi,[1] Yahya Hejazi,[3] Ahmad Laleh,[1] Valiollah Mozaffarian,[4] Shiva Masoudi,[5] and Abdolhossein Rustaiyan[6]

UDC 547.913

The hydrodistilled oils from the aerial parts of Ferula latisecta *and* Mozaffariania insignis, *which is endemic to Iran, were analyzed by GC and GC/MS. (*Z*)-Ocimenone (32.4%), (*E*)-ocimenone (20.3%), and* cis*-pinocarvone (11.4%) were the main components among the 22 constituents characterized in the oil of* F. latisecta, *representing 87.7% of the total components detected. Twenty-five compounds were identified in the oil of* M. insignis, *representing 99.0% of the total oil, with octyl acetate (41.1%),* β*-pinene (30.3%), and* α*-pinene (23.9%) as the main constituents. The essential oils were examined for their potential antimicrobial activities.*

Key words: *Ferula latisecta, Mozaffariania insignis,* Umbeliferae, essential oil composition, antimicrobial activity, (*Z*)-ocimenone, (*E*)-ocimenone.

The genus *Ferula*, which belongs to the Umbelliferae family, is widespread in the Mediterranean area and Central Asia [1]. The flora of Iran comprises 30 species of *Ferula* of which 15 are endemic [2, 3]. The resins of the plants have been used in China as a remedy for malaria and dysentery and also as an insecticide [4]. Other medical uses are also reported, such as sedative, antispasmodic, for toothache, asthma, cough, epilepsy, fever, irritable colon, as an antihysteric, for feminine sterility, and against rheumatism [5–7]. Toxicity in cattle due to *Ferula* is well known as ferulosis, which is a hemorrhagic disease caused by *F. communis* and many other *Ferula* species, mostly due to their 4-hydroxylated and phenylated coumarins [8].

A large number of studies have concerned the genus *Ferula* and reported newly identified compounds extracted by solvent. The reported compounds are mainly coumarin derivatives and sesquiterpene alcohol esters, most of which are aromatic esters bearing the daucane skeleton [9–11].

Few studies have reported the chemical composition of *Ferula* essential oils. Previously, we reported the essential oil composition of *F. stenocarpa, F. macrocolea, F. flabelliloba, F. galbaniflua, F. microcolea,* and *F. hirtella* [12–16].

The present work presents the chemical composition and antibacterial activities of the hydrodistilled oils of *F. latisecta* Rech. f. & All and *M. insignis* Mozaff. of Iranian origin for the first time.

The composition of the oils of *F. latisecta* and *M. insignis* is given in Tables 1 and 2, respectively.

The oil of *F. latisecta* was characterized by large amounts of oxygenated monoterpenes (75.3%), with (*Z*)-ocimenone (32.4%), (*E*)-ocimenone (20.3%), and *cis*-pinocarvone (11.4%) being the major constituents found; smaller amounts of monoterpene hydrocarbons and sesquiterpenes (8.3 and 4.1%, respectively) were also found.

The major constituents of the oils from the aerial parts of *F. stenocarpa* and *F. macrocolea* were *β*-pinene (30.1 and 15.9%) and *α*-pinene(48.8 and 10.4%), respectively [9, 10].

1) Department of Chemistry, Shahid Beheshti University, Eveen, Tehran, Iran, fax 98 21 2240 3041, e-mail: zohre1340@hotmail.com; 2) Department of Phytochemistry, Medicinal Plants and Drugs Research Institute, Shahid Beheshti University, Tehran, Iran; 3) Department of Chemistry, Payame Noor University of Sari, Sari, Iran; 4) Research Institute of Forests and Rangelands, Tehran, Iran; 5) Department of Chemistry, Islamic Azad University, Central Tehran Branch, Tehran, Iran; 6) Department of Chemistry, Science and Research Campus, Islamic Azad University, Tehran, Iran. Published in Khimiya Prirodnykh Soedinenii, No. 6, pp. 561-563, November-December, 2006. Original article submitted November 7, 2005.

0009-3130/06/4206-0689

TABLE 1. Chemical Composition of the Essential Oil from *Ferula latisecta*

Compound	RI	Percentage	Compound	RI	Percentage
α-Thujene	931	0.4	Thuj-3-en-10-al	1181	1.0
α-Pinene*	939	3.6	*cis*-Pinocarveol	1183	11.4
β-Pinene*	980	0.3	Verbenone	1204	0.4
o-Cymene	1022	0.4	(*Z*)-Ocimenone	1231	32.4
p-Cymene*	1026	0.4	(*E*)-Ocimenone	1239	20.3
Limonene*	1031	1.9	Carvone*	1242	0.8
1,3,8-*p*-Menthatriene	1111	1.3	α-Copaene	1376	0.5
(*E*)-Tagetone	1146	3.0	β-Cubebene	1390	1.5
(*Z*)-Tagetone	1153	3.9	Germacrene D	1480	0.5
Isoborneol	1156	1.2	*ar*-Curcumene	1483	1.3
α-Phellandren-8-ol	1166	0.9	Cuparene	1502	0.3

Identification method: RI, MS; * - RI, MS, Co-I.

TABLE 2. Chemical Composition of the Oil of *Mozaffariania insignis*

Compound	RI	Percentage	Compound	RI	Percentage
α-Thujene	931	0.5	*trans*-Carvyl acetate	1337	0.1
α-Pinene	939	23.9	Neryl acetate	1365	0.1
Camphene	953	0.1	Geranyl acetate	1383	Tr.
β-Pinene	980	30.3	Methyl eugenol	1401	0.2
Limonene	1031	1.8	δ-Cadinene	1524	Tr.
(*E*)-β-Ocimene	1050	Tr.	Caryophyllene oxide	1581	Tr.
Linalool	1098	0.3	Hexadecane*	1600	Tr.
trans-Verbenol	1144	0.1	Heptadecane*	1700	Tr.
Terpin-4-ol	1177	0.1	Octyl octanoate	1781	0.3
Octyl acetate	1211	41.1	Octadecane*	1800	Tr.
Piperitone	1252	Tr.	Nonadecane*	1900	Tr.
Linalool acetate	1257	Tr.	Eicosane*	2000	0.1
Lavandulyl acetate	1289	Tr.			

Tr.: trace (<0.1%).
Identification method: RI, MS; * - RI, MS, Co-I.

The oil from the aerial parts of *F. flabelliloba* contained δ-cadinene (13.2%), α-cadinol (12.0%) and cadina-4,1(10)-dien-8β-ol (10.9%), and α-pinene (10.0%), as the major components [11].

The major constituent of the stem and root oils of *F. galbaniflua* was β-pinene (46.4 and 58.8%, respectively) [12]. The oil of *M. insignis* was characterized by high amounts of monoterpene hydrocarbons (56.6%), with β-pinene (30.3%) and α-pinene (23.9%) being the major constituents. Also, a considerable amount of an aliphatic ester, i.e., octyl acetate (41.1%), was found in the oil. The sesquiterpene fraction of the oil was relatively small.

As can be seen in Table 3 the essential oil of *F. latisecta* has no effect on *Pseudomonas aeruginosa* but indicated significant activity with inhibition zones about (10–19 mm) against all Gram-positive bacteria and *Escherichia coli*.

The essential oil of *M. insignis* showed maximum inhibitory activity against *Staphylococcus aureus* and indicated significant activity against *Escherichia coli* and *Bacillus subtilis* but was inactive against *Klebsiella pneumoniae, Pseudomonas aeruginosa*, and *Enterococcus faecalis.*

TABLE 3. Antimicrobial Activity of the Oils

Test Organism	Inhibition zone[a]	
	F. latisecta	*M. insignis*
Bacillus subtilis	15	15
Staphylococcus aureus	14	35
Enterococcus faecalis	19	-
Escherichia coli	13	12
Klebsiella pneumoniae	10	-
Pseudomonas aeruginosa	-	-

[a]Diameter of inhibition zones (mm) including diameter of sterile disk (6 mm).
(-), Inactive; (7-14), moderately active; (>14), highly active.

EXPERIMENTAL

Plant Material. The aerial parts of *F. latisecta* were collected from Hezar Masjed mountains, Province of Khoraasan, Iran, and the aerial parts of *M. insignis*, a newly discovered wild *Mozaffariania* in Iran, were collected in the Lavasan area, East of Tehran, both in June 2003, during the flowering stage.

Voucher specimens were deposited at the Herbarium of the Research Institute of Forest and Rangelands (TARI), Tehran, Iran.

Isolation of the Essential Oils. Aerial parts of *F. latisecta* and *M. insignis* were subjected to hydrodistillation using a Clevenger-type apparatus to produce light yellow oils in 0.4 and 0.6% (w/w) yields, respectively.

Gas Chromatography. GC analysis was conducted using a Thermoquest-Finnigan Trace GC instrument equipped with a capillary DB-1 fused silica column (60 m × 0.25 mm i.d., film thickness 0.25 μm). Nitrogen was used as the carrier gas at the constant flow rate of 1.1 mL/min. The oven temperature was held at 60°C for 1 min, then programmed to 250°C at a rate of 5°C/min, and then held for 10 min. The injector and detector (FID) temperatures were kept at 250°C and 280°C, respectively.

Gas Chromatography/Mass Spectroscopy. GC/MS analysis was carried out on a Thermoquest-Finnigan Trace GC/MS instrument equipped with a DB-1 fused silica column (60 m × 0.25 mm i.d., film thickness 0.25 μm). The oven temperature was raised from 60 to 250°C at a rate of 5°C/min then held at 250°C for 10 min; transfer line temperature was adjusted at 250°C. Helium was used as the carrier gas at a flow rate of 1.1 mL/min with a split ratio of 1/50. Identification of the constituents of each oil was achieved by comparison of their mass spectra and retention indices (RI) with those reported in the literature [17], and those of authentic samples.

Antimicrobial Activity. The antimicrobial activities of the essential oils were evaluated by disc diffusion method using Müeller-Hinton Agar for bacteria with determination of inhibition zones [18]. The antimicrobial activity of the essential oil of *F. latisecta* and *M. insignis* were tested against three Gram-positive and three Gram-negative bacteria, The Gram-positive bacteria included *Staphylococcus aureus* ATCC 25923, *Enterococcus faecalis* ATCC 15753, and *Bacillus subtilis* ATCC 9372, and the Gram-negative bacteria included *Klebsiella pneumoniae* ATCC 3583, *Pseudomonas aeruginosa* ATCC 27852, and *Escherichia coli* ATCC 9763.

ACKNOWLEDGMENT

We would like to acknowledge the financial support provided by the Research Council of Shahid Beheshti University.

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