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Short Communication

Chemical composition of the essential oil from *Ziziphora clinopodioids* Lam. from Iran by means of gas chromatography-mass spectrometry (GC-MS)

A. R. Sardashti[1]*, J. Valizadeh[2] and Y. Adhami[1]

[1]Department of Chemistry, Faculty of Sciences, University of Sistan and Baluchestan, P. O. Box 98167 Zahedan, Iran.
[2]Department of Biology, Faculty of Sciences, University of Sistan and Baluchestan, P. O. Box 98167, Zahedan, Iran.

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The essential oil from aerial parts of *Ziziphora clinopodioids* plant was extracted by hydro distillation with efficiency of 1.86% (w/w). The components were characterized by means of gas chromatography– mass spectrometry (GC/MS) using kovat's retention indices. Thirty-two components of the essential oil which composed 98.18% of the total oil were characterized. The major components were pulegone (61.67%), cis-caran-trans-2-ol (12.66%), 1, 8-cineole (10.23%) and 2-β-pinene (2.16%).

Key words: *Ziziphora clinopodioides*, essential oil, pulegone, (-) cis-caran-trans 2-ol, 1, 8-cineole, 2-β-pinene.

INTRODUCTION

The genus Ziziphora belongs to the family Labiatae consisting of four species that is widespread all over Iran. *Ziziphora clinopodioides* Lam. with the common Persian name "kakuti-e kugi" is an endemic species that grows wild in Iran, Afghanistan, Iraq and Azerbaijan (Talish). *Z. clinopodioids* Lam. is an edible medicinal plant with leaves, flowers and stems that are frequently used as wild vegetable or additive in foods to offer aroma and flavor (Zargari, 1993). The plant is used as stomachic, anti-fever, anti-inflammatory, sedative and flavoring agent in Iranian folk medicine. The chemical composition of the essential oil from *Z. clinopodiodes* growing wild in Bakhteyari province was examined by Sajjadi et al. (2005). Twenty-two components were identified in which pulegone was 53.2%, p-metha-1-en-1-ol was 21.4%, 1, 8-cineole was 10.3% and β-pinene was 1.6% (Sajjadi et al., 2005). Meral et al. (2002) in their research about Ziziphora taurica subsp. clinopodioides in Turkey showed the components which have the higher percentage including pulegone (81.86%), limonene (4.48%) and piperitenone (2.30%). In another research undertaken by Verdian-rizi (2008), *Z. clinopodioides* Lam. was collected from North of Iran and the essential oil from this plant with outcome of 0.9% w/w was extracted and 26 components were recognized including 97.62% of total essential oil. The most important components were pulegone (36.45%), piperitenone (19.12%), and menth-2-en-1-ol (5.31%).

*Corresponding author. E-mail: asardashti65@gmail.com. Tel: 0541-2447816. Fax: 0541-2450995.

MATERIALS AND METHODS

Plant materials

The aerial parts of *Z. clinopoclioides* were collected at the flowering stage from the Taftan area in Baluchestan region Southeastern of Iran in June, 2005. Its scientific name was adopted from the Karaj Herbarium, Institute of Iran (Mozaffarian, 1996).

Isolation of essential oil

The aerial parts (20 g) were dried at 25°C in the shade and subjected to hydro distillation using a Clevenger-type apparatus for 4 h obtained an essential oil which was collected in Hexane-solvent and dried with anhydrous sodium sulphate, weighed and stored at 4°C in the dark until use. The efficiency of extraction was 1.861±0.002 w/w and Relative Standard Deviation (RSD) for four extractions were 3.49 ppt.

Gas chromatography–mass spectrometry (GC/MS) analysis

The analysis of the essential oils was performed using a Hewlett-

Table 1. Chemical composition of essential oil from *Ziziphora clinopodioides* Lam.

Compounds	Retention indices	Percentage (%)
Phellandrene	927	0.09
α-Pinene	934	1.47
Camphene	948	0.46
B-D-xylopyranose	968	0.05
Sabinene	974	1.36
2-β-Pinene	976	2.16
β-myrcene	990	0.74
Ethyl-amyl carbinol	997	0.24
Cyclohexene,1-methyl-4-(1-methylidene)	1015	0.08
1,8 -cineole	1034	10.23
Trans-ocimene	1044	0.3
Γ-Terpinene	1058	0.15
(IR)-(+)-Trans-is limonene	1070	1.03
α-Terpinolene	1084	0.33
Cyclopropane, pentyl	1130	0.14
(-)Cis-Caran-trans-2-ol	1155	12.66
Cis-Cyclohexanone-5-methyl-2-(1-methyl ethyl)	1164	0.34
Isopulegone	1175	1.78
3-Cyclohexene-1-methanol / /α/ α4 / - trimethyl	1206	0.21
Pulegone	1253	61.67
Cyclohexene -1-one/3-methyl-6(1-methylethyl) 2-	1262	0.07
Z-Citral	1274	0.05
6-Octen-1-ol/3/7-dimethyl-formate	1279	0.05
Bicyclo[2/2/1 hetan-2-ol/1/7/7-trimethyl-acetate(15-endo)	1286	0.11
1/1-Dimethyl-3-methylidene-2-vinylcyclo hexane	1303	0.18
phenol/4-ethenyl-2-methoxy	1316	0.4
2-Cyclohexan-1-one/3-methyl-6-(1-methylethylidene	1341	1.09
Cyclohexanone/2/5-dimethyl-2-(1-methyletheny)	1349	0.18
Trans-Caryophyllene	1413	0.18
Germacrene D	1475	0.06
(-)Caryophyllene oxide	1576	0.3
(3R*/4S*)-3-(2-Nitro-4-methoxyphenyl)-4-(4-hydroxphenyl)hexane	2144	0.02

Compounds presented in order of elution from the HP-5Ms capillary column; retention index relative to n-alkanes on the HP-5Ms capillary column; percentage based on FiD peak area normalization.

Packard 6890 network GC system, equipped with a 60 m* 0.25 mm id, 0.25 µm HP-5 Ms capillary column, and a HP 5973 mass selective detector. Helium was the carrier gas at 1 ml/min. Injector and MS transfer line temperatures were 250 and 260°C respectively. Column temperature was set at 40°C for 1 min, and then programmed from 40 to 250°C at a rate of 3°C/min, and finally held isothermally for 20 min for GC/MS detection. An electron ionization technique was used with ionization energy of 70 eV; retention indices were calculated by using the retention times of C8 to C26 n-alkenes which were injected into the oil at the same chromatographic conditions based on Vanden's method (Van den Dool and Kartz, 1963).

Identification of compounds

The linear retention indices for all the compounds were determined by co-injection of the sample with a solution containing the homologous series of C8 to C26 n- alkanes. The individual constituents were identified by their identical retention indices, referring to known compounds from the literature (Adams, 1995) and also by comparing their mass spectra with either the known compounds or with the Wiley 7 mass spectral database.

RESULTS AND DISCUSSION

The identified components and their percentages are listed according to their elution order (Table 1). Identification of the components was based on GC retention indices (Vanden and Kartz, 1963). The fragmentation pattern of the mass spectra was computer-matched with Wiley GC/MS library and compared with those reported in other literature s (Adams, 1995). As

Table 2. Chemical composition of essential oil of Z*. clinopodioides* Lam by chemical class.

Chemical composition by chemical class	Percentage (%)	Number of compounds
Monoterpene hydrocarbons	8.17	11
Oxygenated monoterpenes	88.1	9
Sesquiterpene hydrocarbons	0.24	2
Oxygenated sesquiterpenes	0.30	1
Other hydrocarbons compounds	0.32	2
Other oxygenated compounds	1.05	7
Total monoterpenoids	96.2 7	20
Total sesquiterpenoids	0.54	3
Total	98.18	32

shown in Table 2, 32 components which composed 98.18% of the total oil were identified. The major components were pulegone (61.67%), cis-caran trans-2-ol (12.66%), 1, 8-cineole (10.23%) and 2- β -pinene (2.16%). A qualitative comparison of the oil constituents of *Z. clinopodioides* with those of other species reported showed varying compositions. The results of this investigation indicated that the essential oil of aerial parts of *Z. clinopodioides* Lam. was characterized by high contents of oxygenated monoterpenes (88.10%) according to Meral et al. (2002).

Conclusion

An analysis of the essential oil of *Z. clinopodioides* Lam. by GC/MS showed that the components that have the highest percentages are: pulegone, (-) cis-caran-trans-2-ol and 1, 8-cineole. These compounds have anti-bacterial properties and can be used in making anaesthetic and antiseptic drugs. Other important compounds are α-pinene which is found only in the essential oil as a result of strong light influence.

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