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Composition of essential oil and fatty acids of *Centaurea pichleri* ssp. *pichleri*

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Abstract: In this study, the essential oil of *Centaurea pichleri* ssp. *pichleri* was obtained by hydrodistillation using a Clevenger-type apparatus. GC and GC/MS analyzes of the essential oil from *Centaurea pichleri* ssp. *pichleri* were determined the identification of 48 components representing 86.9% of the oil. With this analysis, the major component was found as hexadecanoic acid (31.4%). Subsequent to this component, other major components were caryophyllene oxide (6.4%), spathulenol (6.2%) and dodecanoic acid (4.5%). In addition to this, fatty acid methyl esters (FAMEs) from *Centaurea pichleri* ssp. *pichleri* were prepared for analyzes of fatty acids. By this test the amount of saturated fatty acid (SFA) was found as 47.79% with major fatty acid was stearic acid (18.64%). The amount of monounsaturated fatty acid (MUFA) was found as 16.88% with major fatty acid was oleic acid (14.20%). The amount of polyunsaturated fatty acid (PUFA) was found as 21.29% with major fatty acid was linoleic acid (15.20%). The results from this work were compared with the previous works in terms of essential oils and fatty acids.

Keywords: *Centaurea pichleri ssp. pichleri*; essential oil; GC; GC/MS; fatty acid

1. INTRODUCTION

In Asteraceae family, *Centaurea* genus is one of the largest genus according to having species. *Centaurea* genus is represented with 192 taxa in Turkey, 114 of which are endemic [1]. Many species of the genus *Centaurea* have traditionally been used for their antirheumatic, diuretic, choleretic, stomachic, astringent, cytotoxic, antibacterial, antipyretic and tonic properties [2, 3]. The essential oil compositions of some *Centaurea* species from Turkey have been investigated. Generally, germacrene D, hexadecanoic acid, caryophyllene and caryophyllene oxide were reported to be the major volatile components in the earlier studies. In *Centaurea* genus, *Centaurea pichleri* ssp. *pichleri* is known as "gelin düğmesi, peygamber çiçeği" in Turkey.

Fatty acid, either saturated or unsaturated, is a carboxylic acid with a long aliphatic chain. Most naturally occurring fatty acids have an even numbered chain of carbon atoms ranging from 4 to 28. Fatty acids that have carbon–carbon double bonds are known as unsaturated fatty

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acids whereas acids without double bonds are known as saturated fatty acids. They differ in chain length as well.

To our knowledge, there are no previous studies on the essential oil and fatty acids of *Centaurea pichleri* ssp. *pichleri.* The analysis of essential oil of some plants from *Centaurea* genus have previously been reported [4-10].

2. MATERIAL and METHODS

2.1. Plant Material and Isolation of Essential Oil

The plant *Centaurea pichleri* ssp. *pichleri* was collected on 12 June 2011 at an altitude of 1450 m in Elazığ, Turkey. The plant was identified by Ugur Cakilcioglu (Elazığ Directorate of National Education, Elazığ, Turkey). Voucher speciments of the plant are deposited in Faculty of Pharmacy, Ege University with the number 1470. For obtaining the essential oil of *Centaurea pichleri* ssp. *pichleri* was done by using the method 'hydrodistillation'. For this, the air dried aerial parts of the plant was subjected to distillation by Clevenger apparatus for 3 h. After that the essential oil of the plant was obtained. This essential oil was stored at +4 ºC until using.

2.2. Oil Extraction and Preparation of Fatty Acid Methyl Esters (FAMEs)

For the analysis of fatty acid of *Centaurea pichleri* ssp. *pichleri,* air-dried and powdered aerial parts of the plant was extracted at 60 °C by Soxhlet extractor, using petroleum ether as a solvent. After oil extraction the solvent was removed by a rotary evaporator.

The extracted oil was esterified to determine the fatty acid composition. The fatty acids in the total lipid were esterified into methyl esters by saponification with 0.5 N methanolic NaOH and transesterified with 14% BF3 (v/v) in methanol [11].

2.3. Gas Chromatography (GC)

Gas chromatography analysis was carried out with an Agilent 6890 N GC system. Temperature of FID detector was 300 °C. Simultaneous autoinjection was done on a duplicate of the same column applying the same operational conditions to obtain the same elution order with GC-MS. The relative percentage amounts of the separated compounds were calculated from FID chromatograms.

Fatty acid methyl esters (FAMEs) were analyzed on a HP (Hewlett Packard) Agilent 6890 N model gas chromatograph (GC), equipped with a flame ionization detector (FID) and fitted to a Supelco SP-2380 Fased Silica capillary column (60 m, 0.25 mm i.d. and 0.2 μm). Injector and detector temperatures were set at 250 ºC and 260 ºC, respectively. The oven was programmed at 140 °C for initial temperature and 5 min for initial time. Thereafter the temperature was increased up to 240 °C at a rate of 3°C/min. The total run time was 41.33 min. For the carrier gas helium was used (1 ml/min). Identification of fatty acids was carried out by comparing sample FAME peak relative retention times. The results were expressed as FID response area in the relative percentages. Each reported result was given as the average value of three GC analyzes. The results are offered as means±S.D.

2.4. Gas Chromatography / Mass Spectrometry (GC / MS)

The GC/MS analysis were carried out with an Agilent 5975 GC-MSD system. Innowax FSC column (60 m × 0.25 mm, 0.25 mm film thickness) was used with helium as carrier gas (0.8 mL/min). GC oven temperature was kept at 60 °C for 10 min and programmed to 220 °C at a rate of 4 °C/min and kept constant at 220 °C for 10 min and then programmed to 240 °C at a rate of 1 °C/min. Split ratio was adjusted at 40:1. The injector temperature was set at 250 °C. Mass spectra were recorded at 70 eV. Mass range was from m/z 35 to 450.

The essential oil components were identified by comparison of Mass spectra with those in Wiley GC/MS Library, Adams Library, MassFinder Library and in Baser Library of Essential Oil Constituents which was built up by genuine compounds and components of known oils. Identification of the essential oil components were carried out by comparison of their relative retention times and their relative retention indices (RRI). The results of analysis are given in Table 1.

3. RESULTS and DISCUSSIONS

Composition of the essential oil of *Centaurea pichleri* ssp. *pichleri* is listed in Table 1 with relative retantion rates (RRI) and percentages. With this analysis 48 components were identified in the essential oil of the plant. These components were represented 86.9 % of the oil. Fatty acid composition of *Centaurea pichleri* ssp. *pichleri* is listed in Table 2.

Table 1. Composition of the essential oil of *Centaurea pichleri ssp. pichleri*

RRI	Component	Percentage
1360	1-Hexanol	0.3
1391	(*Z*)-3-Hexenol	1.2
1400	Nonanal	0.3
1452	1-Octen-3-ol	0.5
1553	Linalool	0.7
1612	β-Caryophyllene	1.0
1664	Nonanol	0.8
1668	(*Z*)-β-Farnesene	0.7
1706	α-Terpineol	0.4
1726	Germacrene D	1.8
1741	β-Bisabolene	0.8
1766	Decanol	0.4
1773	δ-Cadinene	0.4
1838	(*E*)-β-Damascenone	0.4
1868	(*E*)-Geranyl acetone	0.3
1871	1-Undecanol	0.1
1941	α-Calacorene	0.6
1945	1,5-Epoxy-salvial(4)14-ene	1.5
1958	(*E*)-β-Ionone	1.4
1973	Dodecanol	0.3
2008	Caryophyllene oxide	6.4
2037	Salvial-4(14)-en-1-one	0.6
2071	Humulene epoxide-II	1.1
2080	Junenol (=Eudesm-4(15)-en-6-ol)	0.5
2098	Globulol	0.5
2130	Salviadienol	0.5
2131	Hexahydrofarnesyl acetone	1.2
2144	Spathulenol	6.2
2179	3,4-Dimethyl-5-pentylidene-2(5H)-furanone	0.6
2187	T-Cadinol	0.5
2192	Nonanoic acid	0.9
2209	T-Muurolol	0.6
2247	Trans-α-Bergamotol	0.3
2255	α-Cadinol	1.4
2278	Torilenol	0.8
2298	Decanoic acid	0.7
2324	Caryophylla-2(12),6(13)-dien-5α-ol (=*Caryophylladienol II*)	1.0

2369	Eudesma-4(15),7-dien-4β-ol	1.3
2389	Caryophylla-2(12),6-dien-5α-ol (=*Caryophyllenol I*)	1.4
2392	Caryophylla-2(12),6-dien-5β-ol (=*Caryophyllenol II*)	1.3
2500	Pentacosane	0.5
2503	Dodecanoic acid	4.5
2509	Methyl linoleate	0.8
2622	Phytol	3.0
2670	Tetradecanoic acid	2.7
2700	Heptacosane	1.5
2822	Pentadecanoic acid	0.8
2931	Hexadecanoic acid	31.4
	TOTAL	**86.9**

Table 2. Fatty acid composition of *Centaurea pichleri ssp. pichleri*

Fatty acids	*Centaurea pichleri* ssp. *pichleri*
C 6:0 (Caproic acid)	2.09[a]
C 8:0 (Caprylic acid)	5.22
C 14:0 (Myristic acid)	3.09
C 15:0 (Pentadecanoic acid)	1.18
C 16:0 (Palmitic acid)	9.28
C 17:0 (Heptadecanoic acid)	2.01
C 18:0 (Stearic acid)	18.64
C 21:0 (Heneicosanoic acid)	4.20
C 22:0 (Behenic acid)	2.08
∑SFA[b]	47.79
C 18:1 ω9 (Oleic acid)	14.20
C 20:1 ω9 (Gondoic acid)	2.68
∑MUFA[b]	16.88
C 18:2 ω6 (Linoleic acid)	15.20
C 18:3 ω6 (γ-linolenic acid)	6.09
∑PUFA[b]	21.29

[a] Values reported are means ± SD of 3 lots analysed.
[b] SFA: Saturated fatty acids, MUFA: Monounsaturated fatty acids, PUFA: Polyunsaturated fatty acids.

4. DISCUSSION

From the analysis of essential oil of the plant, it is seen that hexadecanoic acid is the major acid. Hexadecanoic acid was previously found as the major component of essential oils of *Centaurea aladagensis, C. luschaiana, C. tossiensis, Centaurea aggregata subsp. aggregata, C. balsamita, C. behen, C. wagenitzii, C. iberica, C. hyalolepis* and *C. polyclada* from Turkey [9, 10, 12, 13]. Hexadecanoic acid, the most common saturated fatty acid which is found in animals, plants and microorganisms was known to raise plasma cholesterol concentrations and also dietary intakes of saturated fatty acids were shown to increase the possibility of coronary heart diseases [14].

With the fatty acid analysis, totally 13 fatty acids were identified in the oil of the plant. For the saturated fatty acids, the major acid was stearic acid as 18.64%. For the monounsaturated fatty acids the major acid was oleic acid as 14.20%. for polyunsaturated fatty acids the major acid was linoleic acid as 15.20%. Saturated fatty acids amounted to 47.79% of the total fatty acids, while the unsaturated fatty acids were 38.17%. There have been previous

studies on fatty acids of some *Centaurea* species [5, 15]. When these results are compared with the previous studies, our results are shown meaningful.

Linoleic acid, for the major polyunsaturated acid, is necessary in adequate amounts for health. Lack of dietary essential fatty acids such as linoleic acid has been implicated in aetiology of diseases including cardiovascular disease and its progression [16]. And also oleic acid, the major monounsaturated fatty acid for the plant, has the capability to lower blood cholesterol levels like linoleic acid. Intake of these fatty acids (oleic and linoleic acids) are promoted by nutritionists and the health professionals [17]. Oleic acid, with the ability of reducing low-density lipoprotein (LDL) levels and possibly increasing high-density lipoprotein (HDL) levels, is known as a monounsaturated fatty acid in normal diet [16,18].

In conclusion, this is the first report on the essential oil and fatty acid composition of *Centaurea pichleri* ssp. *pichleri*.

Conflict of Interests

Authors declare that there is no conflict of interests.

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