



Hydrodistilled Volatile Oil from Stems of *Eryngium creticum* Lam. in the Marginal Brackish Regions of Semnan Province by Using Gas Chromatography Combined with Mass Spectrometry

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The present report deals with chemical composition of the volatile oil from stems of *Eryngium creticum* Lam. a growing wild plant in Iran. All the samples were collected from different brackish and hill slope locations during the flowering stage in Semnan province, Iran. The oil was obtained by using a modified Clevenger-type apparatus and the respective analyses were performed by means of GC and GC-MS instrumentations. It was obtained as a clear yellowish colour liquid involving 0.18 w.w⁻¹ (%). Seventeen components were identified constituting 91.4 % of the oil composition. The major components of the oil were found to be bornyl acetate (28.4 %), camphor (17.8 %), α -pinene (12.1 %), germacrene D (9.4 %), borneol (8.6 %) and α -thujene (4.2 %). Based upon the chemical profile, the essential oil was mainly characterized by the presence of higher amounts of oxygenated monoterpenes.

Key Words: *Eryngium creticum* Lam., GC-MS, Hydrodistillation, Bornyl acetate, Essential oil.

INTRODUCTION

The genus *Eryngium* belongs to Apiaceae family and consists 9 thorny species in Iran. Among them, *E. bungei*, *E. caucasicum* as well as *E. billardieri* are the most common species worldwide¹. The chemistry, genetic diversity and properties of several species of *Eryngium* have been previously studied in detail in the literature²⁻¹¹. However, the essential oils of only few species have been previously reported¹²⁻²¹. Some of the common pharmaceutical and medical applications of the most familiar species of *Eryngium* genus in folk medicine are listed in Table-1. Semnani²² has reported analysis of essential oil of *E. bungei* Boiss. (Umbelliferae) collected from the suburb and coastal margins of Sari, North of Iran. In this work, the chemical profile of hydrodistilled volatile oil was mostly characterized by cuminal alcohol (55.3 %), terpinolene (14.6 %), carvacrol (8.9 %) and limonene (7.5 %).

Nowadays, modern sample preparation in analytical chemistry is characterized by simplification, miniaturization, high enrichment and minimization of sample amount and solvent²³.

The main goal of present study is to operate and optimize a simple and reliable process for the extraction of essential oil by utilization of hydrodistillation method from the fresh stems of *Eryngium creticum* Lam.

TABLE-1
COMMON PHARMACEUTICAL PROPERTIES
OF SOME ERYNGIUM GENUS

<i>Eryngium</i> genus	Applications
<i>E. glaciale</i>	Diuretic and laxative properties to treat urinary ailments, edemas acidosis as well as an aid to digestion
<i>E. caucasicum</i> Trautv.	Enforcing generative power, diuretic, lenitive and appetizer
<i>E. foetidum</i> L.	Food flavouring, traditional medicine for colds, asthma, cough, sinusitis, rheumatism, diarrhea

To the best of my knowledge, it is the first report of the utilization of hydrodistillation as an effective, fast and straightforward method for recognizing the volatile composition of stems of *E. creticum* Lam. Also, chemical composition of the volatile oil from *E. creticum* Lam. in Semnan province (Iran) has not been previously studied.

EXPERIMENTAL

The plant material was collected while wearing polystyrene gloves during the flowering stage in Mahishahr Mountains located in the east of Semnan Province, Iran, in May 2010, at an altitude of 1530 mtrs. Special care was taken to avoid extra damaging and minimizing cross-contamination of plant parts.

Botanical identification: The plant was identified by a local botanist and a voucher specimen was deposited at the Herbarium of the Research Institute of Forests and Rangelands, Tehran, Iran.

Drying and hydrodistillation: Fresh stems of *E. creticum* Lam. were air-dried in the shade at 25 °C for one week, cut into pieces of size over the range 1-4 cm and grounded to a homogeneous powder. The powder was immediately placed in a 2 L round bottom flask containing an adequate amount of distilled water for the plant material to be completely immersed. Afterwards, hydrodistillation was conducted on 150 g portion of the stems powder by using a typical Clevenger-type apparatus to produce oils lighter than water²⁴. The collection of the oil was terminated when no increase in essential oil level was observed for longer than 1 h, with total distillation times between 2 and 3.5 h.

The corresponding pale yellowish oil was recovered in yield of 0.18 % (w/w) and was dried over anhydrous sodium sulfate, weighed and kept in a tightly Teflon-capped amber vial at -10 °C until analysis with a meticulous care.

Chromatographic analysis: Gas chromatography analysis was performed on a Shimadzu 15 A gas chromatograph equipped with split/splitless injector (250 °C) and flame ionization detector (FID) at 280 °C. Nitrogen of high-purity was used as carrier gas at a flow rate of 1 mL/min. A SE 30 column (50 m × 0.2 mm) with film thickness of 0.3 µm was employed for separation of the constituents. The column temperature was kept at 90 °C for 3 min, then increased to 220 °C with a 5 °C/min rate and kept constant for 5 min at final temperature. Relative percentages were calculated using a Shimadzu C-R4A Chromatopac and peak area was taken into consideration.

A Hewlett-Packard 6890/5973 apparatus fitted with a HP-5 MS column (30 m × 0.25 mm, film thickness 0.25 µm) was used. The column temperature was kept at 60 °C for 3 min and programmed to 220 °C at a rate of 5 °C/min and kept constant at 220 °C for five min. Helium was the carrier gas (1 mL/min). The split ratio was 1:50. The MS spectrometer was fitted with an electron impact (EI) source operated at an ionization potential of 70 eV with an ion source temperature of 200 °C and a resolution equal to 1000. All the mass spectra were recorded in the range of m/z. 30-500 amu in the full-scan acquisition mode. The interface temperature was 280 °C. The hexanic solutions of a homologous series of saturated paraffins (C₉-C₂₁) and of essential oils were analyzed in GC-FID and GC-MS, using both columns under the same conditions. The linear retention indices were calculated for all volatile components.

Qualitative and quantitative analyses: Determination and identification of the constituents of the oil were made by comparing their mass spectral fragmentation pattern concerning authentic samples and retention indices (RI) relative to C₉-C₂₁ n-alkanes with those given in the literature²⁵⁻³³ and data stored in a MS library (Wiley 275), as well as matching them with National Institute of Standards and Technology mass spectra library (NISTMSL) package with a resemblance percentage above 85 %. Relative percentage amounts of the components were directly calculated from peak area using a Shimadzu C-R4A Chromatopac on the SE-30 column, considering the

sum of all eluted peaks as a 100 % without the use of correction factor.

RESULTS AND DISCUSSION

The retention indices, contents and total contents of chemical compositions in essential oils obtained from stems of *E. creticum* Lam. are listed in Table-2. Seventeen constituents (91.4 %) were identified in this oil: four monoterpene hydrocarbons (MH = 19.5 %), five oxygenated monoterpenes (OM = 57.3 %), five sesquiterpene hydrocarbons (SH = 12.1 %), two non-terpenoid hydrocarbon (NH = 1.6 %) and one oxygenated diterpene (OD = 0.9 %). Based on this information, oxygenated monoterpenes were the major constituents and a ranking order of the groups of constituents as OM > MH > SH > NH > OD was observed in the volatile flower oil. Bornyl acetate was the most abundant constituent (28.4 %), followed by camphor (17.8 %), α-pinene (12.1 %), germacrene D (9.4 %), borneol (8.6 %) and α-thujene (4.2 %).

TABLE-2
CONSTITUENTS OF ESSENTIAL OILS FROM STEMS OF
Eryngium Creticum LAM. EXTRACTED WITH HD METHOD

No.	Compounds ^a	RI ^b	%
1	α-Thujene	924	4.2
2	α-Pinene	932	12.1
3	Camphene	946	1.7
4	β-Pinene	974	1.5
5	1,8-Cineol	1026	2.1
6	Camphor	1141	17.8
7	Borneol	1165	8.6
8	Cis-pinocarveol	1182	0.4
9	Bornyl acetate	1287	28.4
10	Eugenol	1356	0.5
11	Coumarin	1432	1.1
12	trans-β-Farnesene	1454	0.3
13	γ-Himachalene	1481	0.2
14	Germacrene-D	1484	9.4
15	β-Selinene	1489	0.1
16	α-Selinene	1498	2.1
17	Phytol	1943	0.9
Total			91.4

^aThe compounds have been sorted according to their Kovats retention indices on HP-5MS capillary column; ^bRI: Retention Index reported in literature on DB-5 column

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REFERENCES

1. V. Mozaffarian, A Dictionary of Iranian Plant Names, Farhang Moaser, Tehran, Iran (1996).
2. D. Drake and J. Lam, *Phytochemistry*, **11**, 2651 (1972).
3. K. Hiller, K.Q. Nguyen and P. Franke, *Pharmazie*, **33**, 78 (1978).
4. O.R. Simon and N. Singh, *West Indian Med. J.*, **35**, 121 (1986).
5. Z. Yaniv, A. Dafni, J. Friedman and D. Palevitch, *J. Ethnopharmacol.*, **19**, 145 (1987).

6. S. Al-Khalil, *Alexendria J. Pharm. Sci.*, **8**, 73 (1994).
7. A. Alkofahi, A.J. Sallal and A.M. Disi, *Phytother. Res.*, **11**, 540 (1997).
8. M.T. Saenz, M.A. Fernandez and M.D. Garcia, *Phytother. Res.*, **11**, 380 (1997).
9. M.D. Garcia, M.T. Saenz, M.A. Gomez and M.A. Fernandez, *Phytother. Res.*, **13**, 78 (1999).
10. E.S. Menges and J. Kimmich, *Am. J. Bot.*, **83**, 185 (1996).
11. M. Gaudeul, P. Taberlet and I. Till-Bottraud, *Mol. Ecol.*, **9**, 1625 (2000).
12. P.A. Leclercq, N.X. Dung, V.N. Lo and N.V. Toanh, *J. Essent. Oil Res.*, **4**, 423 (1992).
13. K.C. Wong, M.C. Feng, T.W. Sam and G.L. Tan, *J. Essent. Oil Res.*, **6**, 369 (1994).
14. J.A. Pino, A. Rosado and V. Fuentes, *J. Essent. Oil Res.*, **9**, 123 (1997).
15. J.A. Pino, A. Rosado and V. Fuentes, *J. Essent. Oil Res.*, **9**, 467 (1997).
16. A.P. Martins, L.R. Salgueiro, A.P. da Cunha, R. Vila, S. Canigueral, F. Tomi and J. Casanova, *J. Essent. Oil Res.*, **15**, 93 (2003).
17. M.I. Cobos, J.L. Rodriguez, A. de Petre, E. Spahn, J. Casemiro, A.G. Lopez and J.A. Zygadlo, *J. Essent. Oil Res.*, **14**, 82 (2002).
18. J.J. Brophy, R.J. Goldsack, L.M. Copeland and J. Pala-Paul, *J. Essent. Oil Res.*, **15**, 392 (2003).
19. J. Pala-Paul, J.J. Brophy, R.J. Goldsack, L.M. Copeland, M.J. Perez-Alonso and A. Velasco-Negueruela, *Aust. J. Bot.*, **51**, 497 (2003).
20. F. Sefidkon, M. Dabiri and A. Alamshahi, *J. Essent. Oil Res.*, **16**, 42 (2004).
21. J. Pala-Paul, M.J. Perez-Alonso, A. Velasco-Negueruela, J. Varade, M.A. Villa, J. Sanz and J.J. Brophy, *J. Chromatogr. A*, **1074**, 235 (2005).
22. K. Morteza-Semnani, *J. Essent. Oil Res.*, **17**, 485 (2005).
23. C. Zhang, M. Qi, Q. Shao, S. Zhou and R. Fu, *J. Pharmac. Biomed. Anal.*, **44**, 464 (2007).
24. British Pharmacopoeia HMSO, London, 2: A137-A138 (1998).
25. R.P. Adams, Identification of Essential Oils Components by Gas Chromatography/Quadrupole Mass Spectrometry, Allured Publishing Corporation, Illinois, USA (2001).
26. R.P. Adams, Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry, Allured Publishing Co., Carol Stream, IL, USA (2007).
27. M. Mohammadhosseini, A. Pazoki and H. Akhlaghi, *Chem. Nat. Comp.*, **44**, 127 (2008).
28. H. Akhlaghi, A. Shafaghat and M. Mohammadhosseini, *J. Essent. Oil Bearing Plant*, **12**, 365 (2009).
29. H. Akhlaghi, M.R. Saiidi-Asl and M. Mohammadhosseini, *Chem. Nat. Comp.*, **45**, 448 (2009).
30. M. Mohammadhosseini, A. Pazoki, H.A. Zamani, H. Akhlaghi and M. Nekoei, *J. Essent. Oil Bearing Plant*, **13**, 704 (2010).
31. M. Mohammadhosseini, A. Pazoki, H.A. Zamani and H. Akhlaghi, *J. Essent. Oil Bearing Plant*, **14**, 101 (2011).
32. M. Mohammadhosseini, H.A. Zamani, H. Akhlaghi and M. Nekoei, *J. Essent. Oil Bearing Plant*, **14**, 559 (2011).
33. M. Mohammadhosseini, *Asian J. Chem.*, **24**, 1432 (2012).