Advanced Herbal Medicine, 2017; 3(2): 42-49.

herbmed.skums.ac.ir

Study of phytochemical characteristics *Artemisia persica Boiss* in Ilam Province

Seyed Ata Siadat[1], Fatemeh Direkvand-Moghadam[2*]

[1]Agronomy Dept., Ramin University of Agriculture and Natural Resources, Ahwaz, I.R. Iran; [2]Central Lab Dept., Ramin University of Agriculture and Natural Resources, Ahwaz, I.R. Iran.

Received: 4/Jun/2017 Accepted: 18/Aug/2017

ABSTRACT

Background and aims: The genus *Artemisia* is one of the largest and most widely distributed of the nearly 100 genera in the tribe *Anthemideae* of the *Asteraceae* (*Compositae*). 34 species of *Artemisia* have been reported in Iran. Several secondary metabolites characterize the chemical composition of the genus *Artemisia*. Therefore, the current study aimed to investigate phytochemical Characteristics of *Artemisia persica Boiss* collected from the farm and natural habitat of Ilam.

Methods: The aerial parts of plants were collected from farm and Kabirkooh Mountain. After extraction of Artemisinin, the analysis was performed with an HPLC system. Extraction of essential oil was done by distilled hydro. Phytocomponents identified in *Artemisia persica Boiss* essential oils by GC/MS system.

Results: The essential oil yield was reported in Kabirkooh Mountain and farm 0.92% and 0.6%, respectively. The major oil compounds of *Artemisia persica Boiss* collected samples were included: α-Pinene, 1,8-Cineole, (Z)-Sabinene hydrate, (E)-Pinocarveol, Pinocarvone, Artedouglasia oxide C, Laciniata furanone E, Artedouglasia oxide D, Artedouglasia oxide B. The Artemisinin was 2.7 ppm in the Kabirkooh Mountain sample. However the Farm sample had 1.5 ppm.

Conclusions: To achieve the appropriate level of the target compounds, it is important to consider an appropriate place for sampling.

Keywords: *Artemisia persica Boiss*, Essential oil, Artemisinin, Phytochemical.

Original article

INTRODUCTION

The genus *Artemisia* is one of the largest and most widely distributed of the nearly 100 genera in the tribe *Anthemideae* of the *Asteraceae* (*Compositae*).[1] 34 species of *Artemisia* have been reported in Iran.[2] Chemical composition and biological activities of *Artemisia* spp essential oils have been reported recently.[3]

60 constituents were identified in essential oil from aerial parts, leaves,

[*]**Corresponding author:** Fatemeh Direkvand-Moghadam. Researcher of Central lab Dept., Ramin University of Agriculture and Natural Resources, Ahwaz, I.R. Iran, Tel:00989167108576, E-mail: direkvand.fatemeh@yahoo.com

flowers and roots of *Artemisia persica Boiss* from Iran. The oils were extracted by hydrodistillation and the composition of the oils were analyzed by a combination of GC and GC/MS.[4]

There are several secondary metabolites characterize in the chemical composition of the genus *Artemisia*. A survey of the literature indicates that almost all classes of compounds are present in the genus, with particular reference to terpenoids and flavonoids.[5] Sefidkon et al evaluated the antimicrobial effect of the essential oil of *Artemisia spicigera* against Gram positive and Gram negative bacteria.[6] Their results indicated that all bacteria except Pseudomonas aeruginosa have the oil produced inhibition zone more than 20 mm.

Artemisinin is a sesquiterpene compound which has been isolated from *Aretemisia annua* for the first time.[7] Previous studies have been reported the treatment effects of Artemisinin and its derivatives in Malaria, Hepatitis B, various types of Cancer, especially blood Cancer, and Leishmaniasis.[8-10]

Due to the effect of geographical location, altitude and climate on the yield and composition of volatile oils, and in order to know about phytochemical content variations of *Artemisia persica Boiss* in Ilam Province, Kabirkooh Mountain natural habitat and Farm were selected.[11] The current study was aimed to evaluate the phytochemical characteristics of *Artemisia persica Boiss* collected from Kabirkooh Mountain and Farm.

METHODS

The geographical region where samples were taken is shown in Table 1.

Table 1: Geographical profile the region

Region Name	Latitude	Longitude	Height
Farm	33º 36′	46º 36′	1427 m
Kabirkooh Mountain	33º 09′	47º 24′	3050 m

The aerial parts of plants were collected from Farm and Kabirkooh Mountains. Plant material was identified by Ramin Agriculture and Natural Resources University of Khuzestan and transfered to analytical chemistry laboratory for study of phytochemical characteristics.

Approximately, 5 g of each plant sample was weighed accurately and macerated with 250 mL of *n*-hexane at room temperature for 2 days using a laboratory shaker. Then, the *n*-hexane phases

were filtrated and evaporated under vacuum until dryness. The residue was dissolved again in 100 mL of *n*-hexane and the *n*-hexane phase was washed in a separatory funnel with 2 % NaOH solution to get rid of the impurity. After abandoning the alkali solution present in the lower layer, the upper solution was washed with distilled

water several times until it was neutralized. The extract, obtained after distillation under vacuum at 45 ºC in rotary evaporator, was dissolved with 95% ethanol and then filtrated in 250 mL measuring flask. Then, 10 mL of filter liquor was transferred into a 100 mL measuring flask. 40 mL of 0.2 % NaOH solution was added in the flask, and then, let it react at 50 ºC for 30 min. After that, 0.08 mol/L acetic acid solution was filled up to the mark.[12]

The analysis was performed with an HPLC system consisting of an HPLC Knuer smartline series

quaternary pump with degasser and a photodiode array detector. The Nucleodur C18 column (5 µm; 250 mm x 4.6 mm), at 30 °C. The system was controlled and data analysis was performed with clarity system. All calculations concerning the quantitative analysis were performed with external standardization by measurement of peak areas. A mobile phase consisting of formic acid (%0.2 v/v): acetonitrile (50: 50) by isocratic elution was chosen to achieve maximum separation and sensitivity. Flow rate was 1.0 mL/min. Column temperature was set at 30 °C. The samples were detected at 254 nm using photodiode array detector.[13]

100 gm of cleaned and dried plant material was powdered using metal mortar and pestle and placed in a round bottom flask fitted with condenser hydro distilled for 3 hrs at atmospheric pressure and constant temperature. The strongly aromatic oil was separated from the water layer using n-Hexane and the solvent was removed by boiling.

The essential oils of *Artemisia persica Boiss* were analyzed using a Agilent GC (7890) MS (5975) gas chromatography-mass spectrometer (GC-MS) fused with a capillary column of HP-5 MS (30 m × 0.25 mm i.d., 0.25 µm film thickness) with ionization potential of 70 ev. Helium was used as carrier gas at a constant flow of 0.8 ml/min and an injection volume of 1 µL was employed in injector temperature 290 °C; and Ion-source temperature 280 °C. The oven temperature was programmed from 50°C (isothermal for 5 min), with an increase of 3 °C/min, to 240 °C and with an increase of 15 °C/min, to 280 °C held for 10 min. Phytocomponents identified by comparison of their mass spectra with the Wiley 7n and National Institute of Standards and Technology (NIST5.0) libraries. Kovats indices of components were obtained by the aid of standard n-alkanes (C8-C20) injection, under the same chromatographic conditions.[14]

RESULTS

Essential oil yield was calculated by determining the percentage of moisture to the dry weight of each sample at the time of extraction (Table 2). The retention times and chemical composition of phytocomponents in *Artemisia persica Boiss* essential oil are presented in Table 3.

Table 2: The yield essential oil *Artemisia persica Boiss*

Location	Yield essential oil (%)
Farm	0.6
Kabirkooh Mountain	0.92

Table 3: Phytocomponents identified in *Artemisia persica Boiss* essential oils

Component	KI	Component%	
		Field	Kabirkooh Mountain
α-Thujene	0930	0.08	0.10
α-Pinene	0939	6.66	3.43
Camphene	0954	0.33	0.3
Sabinene	0975	0.3	0.08
ß-Pinene	0979	0.25	0.17
Myrcene	0991	-	0.08
α-Terpinene	1017	0.18	-
p-Cymene	1026	1.8	1.6
1,8-Cineole	1031	5.9	6.3
γ-Terpinene	1060	0.45	0.28
(Z)-Sabinene hydrate	1070	23.12	24.1
(E)-Arbusculone	1071	0.1	-
(E)-Pinocarveol	1139	9.2	10.3
(E)-Verbenol	1141	0.13	0.12
Pinocarvone	1165	7.2	7.9
Borneol	1169	0.9	0.75
Terpineol-4-ol	1177	0.31	0.34
p-Cymen -8-ol	1183	-	0.1
Myrtenal	1196	2.14	2.06
Verbenone	1205	0.09	0.15
(E)-Carveol	1217	0.5	0.41
(E)-Pinocarvyl acetate	1298	0.8	0.92
Phenyl ethyl 3-methyl butanoate	1491	0.05	-
Artedouglasia oxide C	1524	11.9	12.04
Laciniata furanone G	1529	1.37	1.30
Laciniata furanone F	1533	1.86	2.41
Laciniata furanone E	1542	10.08	10.3
Laciniata furanone H	1550	2.08	2.15
Artedouglasia oxide D	1561	4.54	5.15
Artedouglasia oxide B	1582	4.82	5.22

The essential oil yield was reported in Kabirkooh Mountain and field 0.92% and 0.6%, respectively. The major oil compounds of samples of *Artemisia persica Boiss* collected were included: α-Pinene, 1,8-Cineole, (Z)-Sabinene hydrate,

(E)-Pinocarveol, Pinocarvone, Artedouglasia oxide C, Laciniata furanone E, Artedouglasia oxide D, Artedouglasia oxide B. The α-Terpinene, (E)-Arbusculone, and Phenyl ethyl 3-methyl butanoate combinations were only existed in Farm; while, Myrcene and *p*-Cymen-8-ol combinations were only existed in Kabirkooh Mountain samples.

Artemisinin standard chromatogram is presented in Figure 1. The isolation of Artemisinin peak chromatogram of the Artemisinin in extract samples is presented in Figure 2.

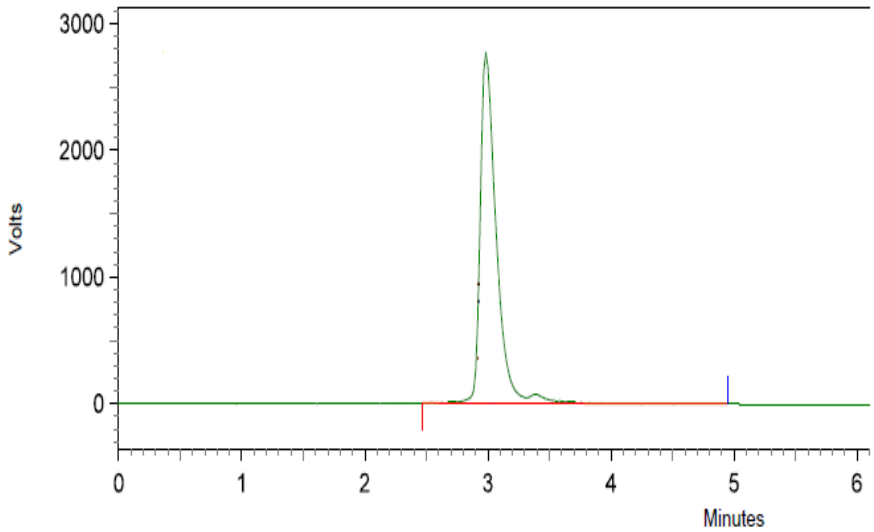

Figure 1: Chromatogram of artemisinin standard

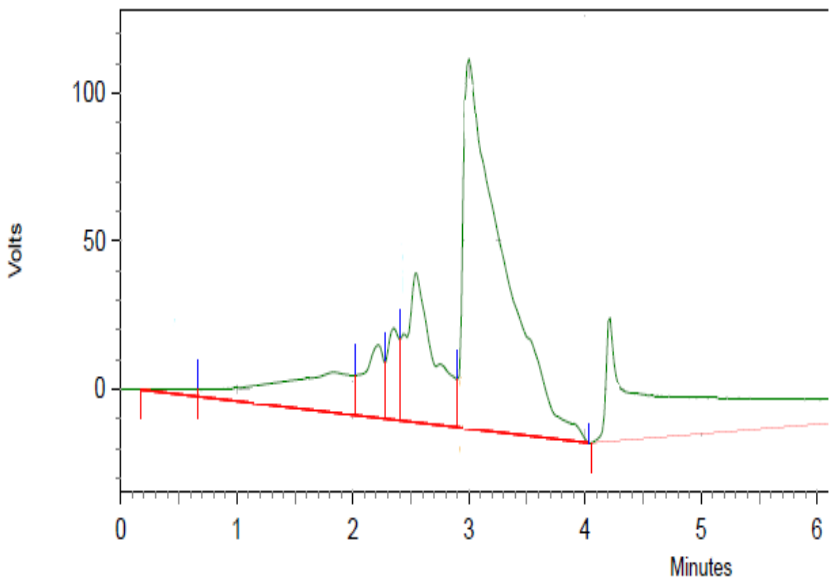

Figure 2: Chromatogram of artemisinin in extract sample

Artemisinin levels were calculated for each sample and presented in Table 4.

Location	Artemisinin (ppm)
Farm	1.5
Kabirkooh Mountain	2.7

The results showed that this concentration is different in samples collected from Farm and Kabirkooh Mountain in Ilam province. The Artemisinin was 2.7 ppm in the Kabirkooh Mountain sample. However, Farm sample had 1.5 ppm.

DISCUSSION

Due to chemical analysis of essential oil *of Artemisia persica Boiss*, main combination of this essential oil includes α-Pinene, 1,8-Cineole, (Z)-Sabinene hydrate, (E)-Pinocarveol, Pinocarvone, Artedouglasia oxide C, Laciniata furanone E, Artedouglasia oxide D, and Artedouglasia oxide B. We found that α-Terpinene, (E)-Arbusculone, and Phenyl ethyl 3-methyl butanoate combinations were only existed in Farm sample, While, Myrcene and *p*-Cymen-8-ol combinations were only existed in Kabirkooh mountain sample. Others studies showed component was 80% monoterpenes and sesquiterpene, but difference was the percentage and type compounds.[15-17] This may be due to season changes, growth stage, collection time of planting, climate conditions and plant growth place.[16,17] Although the productions of secondary metabolites are controlled by genes, but their production will be significantly affected by environmental conditions. Physical and chemical properties of soil, micronutrients and macronutrients are most important factors.[18,19]

The results of the present study showed that the Artemisinin level was higher in the Kabirkooh mountain sample. However, the Farm sample has the lower levels of the Artemisinin. It seems that Artemisinin has been influenced by the growing conditions. Environmental factors play an important role in the production and accumulation of secondary metabolites in medicinal plants. Temperature, precipitation, light intensity, and the above height of sea level are the most important environmental factors affecting the accumulation of secondary metabolites.[20]

CONCLUSION

Temperature, precipitation, light intensity, and the above height of sea level are the most important environmental factors affecting the accumulation of secondary metabolites. To achieve the appropriate level of the target compounds, it is important to consider an appropriate place for sampling. Temperature, precipitation, light intensity, and the above height of sea level are the most important environmental factors affecting the accumulation of secondary metabolites. To achieve the appropriate level of the target compounds, it is important to consider an appropriate place for sampling.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interests.

ACKNOWLEDGMENT

The authors are grateful to thank researchers who generated useful findings for this study.

REFERENCES

1. Mucciarelli M, Maffei M. Introduction to the genus Artemisia. In: Wright CW, Editor. Artemisia. USA: Taylor and Francis; 2002: 1-50.

2. Mozaffarian V. Four new plant species from Ilam province, Iran. Iran J Bot. 2008; 14(2): 87-94.

3. Lopes-Lutz D, Alviano DS, Alviano CS, Kolodziejczyk PP. Screening of chemical composition, antimicrobial and antioxidant activities of Artemisia essential oils. Phytochemistry. 2008; 69(8): 1732-8.

4. Rustaiyan A, Faridchehr A. A review on constituents and biological activities of further Iranian Artemisia species. Int J Pharm Biol Chem Sci. 2014; 3(3): 6-14.

5. Rabie M, Asri Y, Hamzehee B, Jalili A, Sefidkon F. determination of chemotaxonomic indices of artemisia sieberi besser based on environmental parameters in Iran. Iran J Bot. 2012; 18(1): 149-57.

6. Sefidkon F, Tayefeh HE, Fakhari F, Teimouri M. Essential oil composition and antimicrobial activities of oil and alcoholic extract of *Artemisia Spicigera C. Koch* from Mazandaran province. Eco-phytochem J Med P. 2013; 1(2): 1-13.

7. Arab H, Rahbari S, Rassouli A, Moslemi M, Khosravirad F. Determination of artemisinin in Artemisia sieberi and anticoccidial effects of the plant extract in broiler chickens. Trop Anim Health Prod. 2006; 38(6): 497-503.

8. Efferth T, Dunstan H, Sauerbrey A, Miyachi H, Chitambar CR. The anti-malarial artesunate is also active against cancer. Int J Oncol. 2001; 18(4): 767-73.

9. Lai H, Singh NP. Oral artemisinin prevents and delays the development of 7, 12-dimethylbenz [a] anthracene (DMBA)-induced breast cancer in the rat. Cancer Lett. 2006; 231(1): 43-8.

10. Romero MR, Serrano MA, Vallejo M, Efferth T, Alvarez M, Marin JJ. Antiviral effect of artemisinin from *Artemisia annua* against a model member of the Flaviviridae family, the bovine viral diarrhoea virus (BVDV). Planta Med. 2006; 72(13): 1169-74.

11. Mojarrab M, Delazar A, Esnaashari S, Afshar FH. Chemical composition and general toxicity of essential oils extracted from the aerial parts of Artemisia armeniaca Lam. and A. incana (L.) Druce growing in Iran. Res Pharm Sci. 2013; 8(1): 65-9.

12. Hao J-y, Han W, Xue B-y, Deng X. Microwave-assisted extraction of artemisinin from *Artemisia annua L.* Sep Purif Technol. 2002; 28(3): 191-6.

13. Erdemoglu N, Orhan I, Kartal M, Adigüzel N, Bani B. Determination of artemisinin in selected *Artemisia L.* species of Turkey by reversed phase HPLC. Rec Nat Prod. 2007; 1(2-3): 36.

14. Adams RP. Identification of essential oils by gas chromatography quadrupole mass spectroscopy. USA: Allured: Carol Stream, IL; 2001.

15. Bicchi C, Frattini C, Sacco T. Essential oils of three Asiatic Artemisia species. Phytochemistry. 1985; 24(10): 2440-2.

16. Sadeghpour O, Asghari G, Shams Ardekani M. Composition of essential oil of *Artemisia persica Boiss* from Iran. Iran J Pharm Res. 2010; 3(1): 65-7.

17. Mirjalili BF, Meybody MHH, Ardakani MM, Rustaiyan A, Ameri N, Masoudi S, et al. Chemical composition of the essential oil from aerial parts, leaves, flowers and roots of *Artemisia persica Boiss* from Iran. J Essent Oil Res. 2006; 18(5): 544-7.
18. Mahbubi M, Feyzabadi M, Haghi G, Hoseini H. Studying of antimicrobial properties and chemical compound in Oliveria decumbens vent essence. Iran J Med Aro P. 2007; 1(24): 56-65.
19. Palevitch D, editor Recent advances in the cultivation of medicinal plants. VI International Symposium on Medicinal and Aromatic Plants, XXII IHC 208; 1986.
20. Kostova I, Dinchev D. Saponins in tribulus terrestris-chemistry and bioactivity. Phytochem Rev. 2005; 4(2): 111-37.

How to cite the article: Siadat SA, Direkvand-Moghadam F. Study of phytochemical characteristics *Artemisia persica Boiss* in Ilam Province. Adv Herb Med. 2017; 3(2):42-49.