

# **Original Article**

# Essential Oils of *Salvia hydrangea* DC. ex Benth. from Kiasar-Hezarjarib regions, Iran-Impact of eEnvironmental Factors as Quality Determinants

Mahdieh Ebrahimi<sup>1\*</sup> and Somayeh Ranjbar<sup>2</sup>

<sup>1</sup>Department of Range and Watershed Management, Faculty of Soil and Water Science, University of Zabol, Zabol, Iran <sup>2</sup>Department of Range and Watershed Management, University of Zabol, Iran

Article History: Received: 19 February 2016 /Accepted in revised form: 11 June 2016 © 2013 Iranian Society of Medicinal Plants. All rights reserve

# Abstract

The focus of the present study was to investigate the chemical composition of essential oil of Salvia hydrangea DC. ex Benth. growing wild in Kiasar-Hezarjrib, Iran. In addition, effects of altitude and some soil properties on the essential oil quality and quantity of S. hydrangea was identified. The samples collected from two altitudes (1100 and 2000 m) in blossoming period, were analyzed by hydrodistillation method using a clevenger-type apparatus by GC and GC/MS. Ninety-one compounds were identified, representing 99.79% and 99.95% of the oils from the altitudes of 1100 and 2000 m respectively. The main constituents of the oils in altitude 1100 were 1,8-cineole (12.70%), camphor (12.06%), -pinen (11.62%), naphthalene (10.83%), -amorphene (9.30%), bicycleheptan (9.18%) and -cadinene (8.25%). In contrast, in altitude of 2000 m the oil of plant species was rich in naphthalene (19.94%), 1,8-cineole (9.45%), camphor (5.71%), -terpineol (5.11%) and ylangene (4.94%). In both altitudes naphthalene, 1,8-cineole and camphor being the major compounds respectively. Results indicated that the altitude of 2000 m revealed greater quantity. The essential oil compositions showed the highest percentage in the altitude of 1100 m and amount of main and common essential oil compositions was more in the altitude of 1100 m. A high positive correlation was found between the essential oil efficiency and the soil factors of pH, S.P, EC, Mn and Mg in both altitudes while, the common essential oil composition showed negative correlation with these factors. In general, the higher essential oil efficiency and compositions were found in the higher altitude, but percentage of the essential oil compounds was more in the lower altitude therefore, at lower altitudes there are more optimal conditions for taking the S. hydrangea essential oil.

Keywords: Salvia hydrangea DC. ex Benth., Essential oil composition, 1,8-cineole, Camphor;, Naphthalene

# Introduction

Fifty-eight species of the genus *Salvia* (Lamiaceae) are found in Iran, seventeen of which are endemic. Due to the use of this genus or their essential oils in the food, drug and perfumery industries [1-3] we are investigating the oil of *Salvia hydrangea* DC. ex Benth. which grows wild in Iran. The plant species grows in many regions of Iran such as Azerbaijan, Kurdistan, Guilan, Qazvin, Tehran, Kerman, Markazi, Mazandaran, Esfahan, Lorestan, Fars, Sistan and Baluchestan provinces [4].

Despite the great developments in modern pharmaceutical drugs that have saved human beings from different kinds of illnesses, it is impossible to overlook the role of the plants and their positive effects. The active agents present in the plants have always been, and will continue to be, used as irreplaceable substances [5]. Although the growth and the increase in the quantity and quality of the substance in medicinal plants takes place mainly due to genetic processes, environmental factors also play a major role in this regard. Such factors help to bring about certain

<sup>\*</sup> Corresponding author: Faculty of Soil and Water Science, University of Zabol, Zabol, Iran Email Address: maebrahimi2007@uoz.ac.ir

changes in the growth of medicinal plants, as well as changes in the quantity and quality of the active substances [5,6]. Some studies have reported the influence of mineral nutrients [7,8], drought [9], light intensity and altitude [10,11] on the plant growth and essential oil content. These factors can also trigger abrupt activation of qualitative changes in secondary metabolite production [5,12,13]. Since plants cannot escape from the environmental extremes of light, temperature, and drought, nor move to regions with better nutritional conditions, they have thus evolved highly complex mechanisms to integrate physiology and metabolism in order to adapt to the conditions to which they are exposed [14].

Since the amount and quality of a medicinal plant's ingredients in various habitats and areas change due to fluctuations in the plant metabolic activities under different environmental factors, so the quantitative and qualitative of medicinal plants' compounds are different in various countries of the world [6]. Marked variations in the chemical composition of *S. hydrangea* oil from various areas have been reported [15-18].

Nevertheless, ecological factors undoubtedly influence the essential oil content of *S. hydrangea* The present study attempts to investigate the chemical composition of *S. hydrangea* from Mazandaran province, Iran and identify the altitude and edaphic factors that influence the quantity and quality of the essential oil of this plant species as well as the optimal altitude to produce the greatest amount of active substances.

#### **Material and Methods**

#### Plant and Soil Sampling

In this study, the aerial parts of S. hydrangea were collected during the blossoming period from two natural areas (Kiasar and Hezarjarib) located in Mazadaran Province (36° 13' 34" N, 52° 31' 54" E) at two altitudes of 1100 and 2000 m, in May 2014. Surveys of the plant associations in wild habitats were done along transects (100 m) within quadrants (4 m<sup>2</sup> each) with systematically-randomized method. In each altitude, five transects were established in two directions of the general slope. Small quantities (~100-200 g fresh weight/quadrant) of flowering shoots were collected from each surveyed quadrant. The aerial parts of the plant species were dried in the shade (at room temperature).

Soil samples from the root zone were collected from each surveyed quadrat. The soils were put in plastic bags with label; they were thereafter air dried, ground to pass through a 2 mm sieve and taken to the laboratory at the Department of Range and Watershed management, University of Zabol, for analysis of soil physical and chemical properties. The soil's texture was determined using laser diffractometry; [19] pH was determined in a 1:5 soil to distilled water slurry after one hour of agitation using a digital pH-meter (Model 691, Metrohm AG Herisau Switzerland); [20] electrical conductivity (ECe) using an EC-meter (DDS-307, Shanghai, China); [21] organic matter content were determined using the methods described by Lo et al. [22] Available phosphorus was determined by the method of Bray and Kurtz [23]. Potassium was measured by flame photometry method; [24] saturation percentage (SP) determined by weighing method [25]. Mg, Fe, Cu, Zn and Mn determined by atomic absorption method using ICP/OES (GBC Avanta, Australia); [26] Calcium carbonate was determined volumetrically by a calcimeter [27].

## Essential Oil Analysis

Dried aerial parts of the plants (70 g) were chopped in small pieces and submitted to hydrodistillation for 2h using a clevenger-type apparatus with a water-cooled receiver, to reduce hydrodistillationoverheating artifacts. The oils were obtained using n-pentane as a collecting solvent and subsequently they were dried over anhydrous sodium sulphate and stored under N<sub>2</sub> atmosphere in amber vials at 4 °C until they were analyzed. All oils were analyzed within 24 h of their production. Considering the moisture percentage, the essential oil efficiency was measured in dry weight (w/w).

## Gas Chromatography (GC)

Gas chromatographic analysis was carried out on a Perkin-Elmer 8500 gas chromatograph with FID detector and a DB-5 capillary column ( $30 \text{ m} \times 0.25 \text{ mm}$  i.d., film thickness 0.25 µm). The operating conditions were as follows: carrier gas, helium at a flow rate of 2 ml/min; the oven temperature was programmed to 60 °C for 4 min, then 60–220 °C at 4 °C/min; injector and detector temperatures, 240 °C. The percentage composition of the oils was computed in each case from GC peak areas without the use of correction factors.

Ebrahimi and Ranjbar

GC-MS Analysis and Identification of Components

A Hewlett-Packard series 6890 instrument was used, with a DB-5 capillary column (30 m × 0.25 mm i.d., film thickness 0.25  $\mu$ m), programmed as follows: 60 °C for 5 min, then 60–220 °C at 4 °C/min. The carrier gas was helium at a flow rate of 2 ml/min. The components of the oils were identified by their retention time relative to C9– C28 n-alkanes, computer matching with the Wiley 275.L library and as well as by comparison of their mass spectra with those of authentic samples or with data already available in the literature [28]. Statistical analysis

The statistical processing was mainly conducted by T-test. Before performing analysis, data were checked for their normality with the Kolmogorov– Smirnov test and for homogeneity of variance with the Levene's test (p<0.05), and where necessary, data were log-transformed. A probability of 0.05 or lower was considered significant. Correlation coefficients between the essential oil efficiency and composition with the soil properties were also calculated through the Pearson's r coefficient. All statistical calculations were performed using SPSS release 18.0.

# Results

# Essential Oil Composition

The percentage composition of the oils is reported in Table 1 in order of their elution from the DB-5MS column. Twenty-four components were identified for S. hydrangea in altitude of 1100 m constituting 99.97% of the total oil while in altitude of 2000 m, 90 components were identified in the oil of the plant species representing 99.95% of the total oil. In particular, the oil of S. hydrangea in altitude of 1100 m was dominated by 1,8-cineole (12.70%), camphor (12.06%), -pinen (11.62%), naphthalene (10.83%), -amorphene (9.30%), bicycleheptan (9.18%) and -cadinene (8.25%). In contrast, in altitude of 2000 m the oil of S. hydrangea was rich in naphthalene (19.94%), 1,8cineole (9.45%), camphor (5.71%), -terpineol (5.11%) and ylangene (4.94%). Totally 91 compositions were found in both altitudes, but in different percentages. Some components of S. hydrangea exist in just one of the altitude, which can be seen in the table. In both altitude naphthalene, 1,8-cineole and camphor being the major compounds respectively.

Comparison of the Extraction Efficiency and the Essential Oil Composition

The essential oil efficiencies of S. hydrangea collected from two altitudes are shown in Table 2. It was found that the altitude factor had considerable influence on the essential oil efficiency (p<0.01). Significant difference in amount of common and main essential oil compositions of S. hydrangea occurred in the natural habitats (Table 2). Although the amount naphthalene (14.44%) was more in habitat of 2000 m, the highest content of 1,8-cineole (12.66%), pinen (11.40%), camphor (11.20%), bicycloheptan (8.75%) and -cadinene (8.04%) was found in the habitat of 1100 m. In general, in higher altitude the efficiency of essential oil was more but the percentage of the essential oil composition varied in the opposite direction.

# Soil Properties and Correlations

General properties of the soil samples were collected from the natural habitats are given in Table 3. The habitats differed considerably in chemistry. The soil of the habitat in altitude of 1100 m characterized as loamy sand while, in the altitude of 2000 m the soil texture was loamy clay. Soil acidity in both habitats was near neutral (pH=7.86-7.87). Two habitats differed significantly in EC (p=0.05), organic matter (p=0.05), CaCO<sub>3</sub> (p=0.00) P (p=0.01), Fe (p=0.01) and K (p=0.00), so that these factors decreased with altitude, while S.P (p=0.00), Mg (p=0.00) and Mn (p=0.00) varied in the opposite direction. However, there was no significant difference between content of Zn and Cu in two habitats; these factors were more in the altitude of 1100 m.

Results of Table 4 indicted that in the altitude of 1100 m, there was a strong positive relationship between the essential oil efficiency and the soil factors of pH, S.P, EC, Mn and Mg. It was found that essential oil efficiency increased with these While, the common essential factors. oil compositions showed strong negative relationship with the soil factors of pH, S.P, EC, Mn and Mg. Bicycloheptan, naphthalene, ylangene and 1,8cineole were positively related to the content CaCO<sub>3</sub>, OM, P and K. -pinen showed the highest positive relationship with CaCO<sub>3</sub>, OM, P, K, Zn, Fe and Cu. Camphor and -cadinene had the highest positive correlation with Zn (Table 4).

Compound	% in sample		Retention time	
	Hezarjarib (2000 m)	Kiasar (1100 m)		
2-hexenal	0.11	Tr	5.31	
Tricyclene	0.79	1.50	7.24	
-pinene	3.89	6.20	7.74	
-pinene	3.04	11.62	9.34	
-myrcene	1.54	Tr	9.69	
(+)-2-carene	0.08	Tr	10.49	
1,8-cineole	9.45	12.70	11.14	
1,3,6-octatriene, 3,7-dimethyl	0.08	Tr	11.91	
-terpinene	3.38	5.55	12.29	
- terpinolene	1.51	3.21	13.11	
Camphor	5.71	12.06	15.34	
Borneoll	3.43	5.14	16.51	
-terpineol	5.11	Tr	17.45	
Myrtenol	0.07	Tr	17.60	
1-cyclohexene-1-carboxaldehyde,	0.27	0.56	17.99	
Benzene, 1-ethyl-3-(1-methylethy	0.64	Tr	18.37	
2-cyclohexen-1-one, 2-methyl-5-(	0.06	0.90	19.65	
Bicyclo[2.2.1]heptan-2-ol, 1,7,7	3.75	9.18	20.09	
- 3-carene	0.18	Tr	20.68	
Tricyclo[2.2.1.0(2,6)]heptane-3	0.04	Tr	20.75	
Phenol, 2-methyl-5-(1-methylethy	0.67	1.17	20.88	
Myrtenyl acetate	0.11	Tr	21.28	
Azulene, 1,2,3,5,6,7,8,8a-octahy	0.17	Tr	21.53	
3-allyl-6-methoxypheno	0.37	Tr	22.38	
Ylangene	4.94	6.99	22.80	
-cubebene	1.02	3.53	23.21	
Cis-jasmone	0.92	1.90	23.65	
1h-cycloprop[e]azulene, 1a,2,3,4	0.22	Tr	23.91	
Naphthalene, 1,2,3,4,4a,5,6,8a-o	14.88	10.83		
-		2.16	24.31	
-caryophyllene	1.91		25.41	
1h-cycloprop[e]azulene, 1a,2,3,4 -cadinene	1.08	Tr	26.47	
	2.65	8.25	27.06	
Naphthalene, 1,2,3,4,4a,7-hexahy	1.09	Tr	27.82	
Neoalloocimene	1.34	Tr	27.93	
-calacorene	4.45	3.56	28.10	
-selinene	1.06	1.88	28.35	
-crene b	0.09	Tr	28.41	
+ calarene	0.19	Tr _	28.92	
-selinene	0.55	Tr	29.53	
(+)guaiene	0.19	Tr	29.64	
1(5),3-aromadenedradiene	0.43	Tr	30.03	
Fonenol	1.50	2.56	30.15	
Naphthalene, 1,2,3,5,6,8a-hexahy	3.58	Tr	30.34	
Taucadinol	1.03	1.40	30.72	
-eudesmol	3.08	5.22	31.11	
Naphthalene, 1,6-dimethyl-4-(1-m	0.24	Tr	31.60	
O-menth-8-ene	2.46	Tr	29.92	
(3S,4R,5S,6R,7S)-aristol-9-en-3	0.17	Tr	31.71	
1h-cycloprop[e]azulene, decahydr	0.04	Tr	32.02	
Junipercamphor	0.15	Tr	32.14	
Hexadecanoic acid, methyl ester	0.06	Tr	37.77	
Bicyclo[3.1.1]hept-3-ene-2-spiro	0.05	Tr	37.95	

**Table 1** The percentage composition of the essential oils of Salvia hydrangea DC. ex Benth.at two altitudes of Mazandaran.

Dibutyl phthalate	0.20	Tr	38.67
Cisbisabolene	0.07	Tr	38.94
Nona-2,3-dienoic acid, ethyl ester	0.03	Tr	39.39
Cembrene	0.13	Tr	39.88
Phenanthrene, 1,2,3,4,4a,9,10,10	0.20	Tr	40.89
2-methyl-7-exo-vinylbicyclo[4.2	0.05	Tr	41.15
9,12,15-octadecatrienoic acid, m	0.08	Tr	41.82
Phytol	0.45	Tr	42.13
Tricyclo[7.9.0.0(10,18)]octadec	0.08	Tr	42.43
(Z)6,(Z)9-pentadecadien-1-ol	0.10	Tr	42.78
2-ethylhexyl trans-4-methoxycinn	0.03	Tr	43.30
Pimara-8(9),15-diene	0.08	Tr	44.34
5,5'-bis(acetyl)-3,3'-biisoxazol	0.04	Tr	44.79
Longifolen-v2	0.04	Tr	44.79
Ferruginol	0.08	Tr	46.74
5,7-dimethoxy-1-naphthol	0.14	Tr	47.97
-amorphene	Tr	9.30	48.66
1,2-benzenedicarboxylic acid, 3	0.05	Tr	51.12
Nonadecane	0.05	Tr	55.13
Total	99.95	99.79	-

\*Tr: trace

Table 2 Comparison of the essential oil efficiency and common and main components in the habitat sites.

Altitude (m)	Essential oil efficiency	Camphor	-pinen	Naphthalene	Bicycloheptan	-cadinene	1,8-cineole	ylangene
1100	$0.62\pm0.01^{b}$	11.92±0.17a	11.40±0.16a	10.58±0.12b	8.75±0.10a	8.04±0.10a	12.66±0.13a	6.99±0.10a
2000	$1.01{\pm}0.01^a$	5.53±0.12b	2.28±0.71b	14.44±0.26a	3.54±0.18b	2.53±0.18b	9.40±0.22b	4.74±0.12b
Sig	$0.00^{**}$	$0.00^{**}$	$0.00^{**}$	$0.05^{*}$	$0.00^{**}$	$0.00^{**}$	0.05*	$0.05^{*}$

Values ( $\pm$ SE) within a row followed by the different letter are significantly different according to the T-test (p < 0.05). \* *and* \*\*significant at the 0.05 and 0.01 probability level respectively.

Table 3 Characteristics of soil in the habitats of Salvia hydrangea DC. ex Benth.

Soil properties	Altitude (m)		
	1100	2000	Sig
EC (dS/m)	0.68±0.00a	0.43±0.00b	$0.05^{*}$
pН	7.86±0.20a	7.87±0.20a	$0.24^{ns}$
OM (%)	3.00±0.01a	1.01±0.00b	$0.05^{*}$
$CaCO_3(\%)$	49.00±2.60a	19.00±1.13b	$0.00^{**}$
S.P (%)	42.01±1.60b	60.00±2.53a	$0.00^{**}$
P (ppm)	11.50±0.60a	7.30±0.24b	0.01**
K (ppm)	509.00±22.00a	278.00±5.78b	$0.00^{**}$
Mg (ppm)	216.00±5.03b	426.00±18.76a	$0.00^{**}$
Fe (ppm)	7.40±0.22a	3.90±0.01b	0.01**
Mn (ppm)	3.70±0.01b	8.50±0.23a	$0.00^{**}$
Zn (ppm)	1.50±0.02a	1.20±0.01a	0.07 <sup>ns</sup>
Cu (ppm)	0.80±0.00a	0.70±0.00a	$0.60^{ns}$
Texture	Loamy sand	Loamy clay	-

Values ( $\pm$ SE) within a row followed by the different letter are significantly different according to the T-test (p < 0.05). \*significant at the 0.05 probability level, \*\*significant at the 0.01 probability level, n.s means non-significant

Soil	Ylangene	1,8-	-Cadinene	Bicycloheptan	Naphthalene	-pinen	Camphor	Essential
properties		cineole						oil
								efficiency
S.P (%)	-0.98**	-0.97**	-0.94*	-0.98**	-0.97**	-0.99**	-0.95*	0.99**
EC(dS/m)	-0.99**	-0.99**	-0.93*	-0.99**	-0.99**	-0.98**	-0.94*	0.99**
pН	-0.98**	-0.97**	-0.94*	-0.98**	-0.97**	-0.99**	-0.95*	0.99**
$CaCO_{3}(\%)$	$0.98^{**}$	$0.97^{**}$	$0.90^{*}$	$0.98^{**}$	$0.97^{**}$	$0.99^{**}$	$0.95^{*}$	-0.99**
OM (%)	$0.98^{**}$	$0.97^{**}$	$0.93^{*}$	$0.98^{**}$	$0.97^{**}$	$0.99^{**}$	$0.95^{*}$	-0.99**
P (ppm)	$0.98^{**}$	$0.97^{**}$	$0.93^{*}$	$0.98^{**}$	$0.97^{**}$	$0.99^{**}$	$0.95^*$	-0.99**
K (ppm)	$0.98^{**}$	$0.97^{**}$	$0.94^{*}$	$0.98^{**}$	$0.97^{**}$	$0.99^{**}$	$0.95^{*}$	-0.99**
Mg (ppm)	-0.98**	-0.97**	-0.94*	-0.98**	-0.97**	-0.99**	-0.95*	0.99**
Fe (ppm)	$0.81^{*}$	$0.95^{*}$	$0.90^{*}$	$0.81^{*}$	$0.95^{*}$	$0.99^{**}$	$0.93^{*}$	-0.99**
Mn (ppm)	-0.98**	-0.97**	-0.94*	-0.98**	-0.97**	-0.99**	-0.95*	0.99**
Zn (ppm)	$0.98^{**}$	$0.80^{*}$	$0.99^{**}$	$0.98^{**}$	$0.80^{*}$	$0.99^{**}$	$0.97^{**}$	-0.99**
Cu (ppm)	$0.98^{**}$	$0.80^{*}$	$0.80^{*}$	$0.98^{**}$	$0.80^{*}$	$0.99^{**}$	$0.95^{*}$	-0.99**

**Table 4** Pearson's correlation coefficients between soil properties, common essential oil composition, and essential oil efficiency in the habitat of 1100 m.

\*significant at the 0.05 probability level, \*\*significant at the 0.01 probability level

**Table 5** Pearson's correlation coefficients between soil properties, common essential oil composition, and the essential oil efficiency in the habitat of 2000 m.

Soil properties	Ylangene	1,8-	-	Bicycloheptan	Naphthalene	-pinen	Camphor	Essential oil
		cineole	cadinene					efficiency
S.P (%)	-0.84*	-0.84*	-0.85*	-0.97**	-0.84*	-0.99**	-0.97**	$0.97^{**}$
EC(ds/m)	-0.83*	-0.83*	-0.85*	-0.91*	-0.74*	-0.95**	-0.91*	$0.92^*$
pH	-0.84*	$-0.84^{*}$	-0.85*	-0.97**	-0.84*	-0.99**	-0.97**	$0.97^{**}$
CaC0 <sub>3</sub> (%)	0.63 <sup>ns</sup>	0.63 <sup>ns</sup>	$0.85^*$	$0.97^{**}$	0.63 <sup>ns</sup>	$0.98^{**}$	$0.97^{**}$	-0.97**
OM (%)	0.60 <sup>ns</sup>	0.60 <sup>ns</sup>	$0.87^{*}$	$0.96^{**}$	0.70 <sup>ns</sup>	$0.97^{**}$	$0.97^{**}$	-0.97**
P (ppm)	$0.62^{ns}$	$0.62^{ns}$	0.80 <sup>ns</sup>	$0.96^{**}$	0.71 <sup>ns</sup>	$0.98^{**}$	$0.95^{**}$	-0.97**
K (ppm)	$0.70^{ns}$	0.70 <sup>ns</sup>	0.80 <sup>ns</sup>	$0.97^{**}$	0.63 <sup>ns</sup>	$0.98^{**}$	$0.97^{**}$	-0.97**
Mg (ppm)	-0.85*	-0.85*	$0.80^{*}$	-0.97**	$0.60^{ns}$	-0.99**	-0.98**	$0.97^{**}$
Fe (ppm)	$0.92^{**}$	$0.92^{**}$	$0.88^{*}$	$0.99^{**}$	$0.95^{**}$	$0.98^{**}$	$0.96^{**}$	-0.98**
Mn (ppm)	-0.85*	-0.85*	-0.85*	-0.97**	-0.85*	-0.98**	-0.97**	$0.97^{**}$
Zn (ppm)	0.64 <sup>ns</sup>	0.64 <sup>ns</sup>	0.72 <sup>ns</sup>	$0.96^{**}$	$0.80^{ns}$	$0.98^{**}$	$0.95^{**}$	-0.97**
Cu (ppm)	0.60 <sup>ns</sup>	0.60 <sup>ns</sup>	0.73 <sup>ns</sup>	0.91*	0.74 <sup>ns</sup>	$0.95^{*}$	0.91*	-0.92**

\*significant at the 0.05 probability level, \*\*significant at the 0.01 probability level, <sup>n.s</sup> means non-significant

The results of soil properties' correlation associated with the essential oil efficiency in the altitude of 2000 are presented in Table 5. Result showed that there was a strong positive relationship between the essential oil efficiency and the soil factors of pH, S.P, EC, Mn and Mg while, the common essential oil composition showed negative correlation with these factors. Although essential oil efficiency had the highest negative correlation (-0.98) with Fe, cadinene, 1,8-cineole, ylangene, bicycloheptan and naphthalene had the maximum positive correlation with Fe. Camphor had the highest positive correlation (97%) with the factors of CaCO<sub>3</sub>, OM and K. -pinen related positively to CaCO<sub>3</sub>, P, K, Fe and Zn (Table 5).

# Discussion

The present study investigated the chemical composition of the essential oils of *S. hydrangea* and the effects of altitude and soil properties on the essential oil content and composition. Totally, ninety-one components were identified for *S. hydrangea* in the habitat sites. In general, the altitude of 2000 m showed higher essential oil composition, while the altitude of 1100 m indicated lower amount, and as the altitude increased, the essential oil compositions rose.

Environmental factors quantitatively affect the plant's metabolic processes through their effects on the plant development, growth rates and partitioning of assimilates into vital metabolites. External factors often have an especially large influence on the biosynthetic levels and quality of secondary metabolites in the plants [29].

It is important to know the factors that influence the plant production, and to know, for each particular case, their specific requirements. The factors include: (a) physiological variations; (b) environmental conditions; (c) geographic variations; (d) genetic factors and evolution; (e) political/social conditions; and also (f) amount of plant material/space and manual labour needs [30]. Medicinal plants are grown and produced in different ecosystems and sites under the influence of different potential factors, including the altitude as one of the vital determinants in the quantity and quality of the plants [5]. Altitude strongly influences the synthesis and accumulation of the essential oil compositions [11] and it has a vital role in the growth and production of the medicinal plants in a variety of natural ecosystems and areas [31-32, 5]. So that, increasing and decreasing of altitude level can change the temperature, relative humidity, wind speed, available water to the plant's root and sunlight rates, hence, regarding the altitudinal level changes, ecophysiological reactions of the plant will also change [33]. According to Senatore et al. [34] various factors, both endogenous and exogenous, can affect the composition of the essential oil (S. glutinosa); the time of flowering, together with geographical and climatic factors, is important in determining the composition of the oil.

In an investigation of the effect of environmental factors on *Cymbopogon olivieri* (Boiss.) Bor (Poaceae) in four regions of Iran, Sarbaz, Jiruft, Dezfool, and Masjid Soleiman, Mirjalili *et al.* [35] concluded that the nearby altitudes of 300-600 m in the regions of Masjid Soleiman and Jiruft had a greater effect on the function of the essential oil in the lemongrass. The identification of the chemical composition of *Ziziphora clinopodioides* Lam. (Lamiaceae) in different altitudes in several areas indicated that there were different composition percentages at different altitudes and areas, and that other environmental factors, such as soil, also had a role in the differences [36].

As shown in the results of this study, the correlation between the soil properties and the quantity and quality of the essential oil were different. It is interesting to note that in comparison of both habitats, amounts of common essential oil compositions of *S. hydrangea* were more in Kiasar (altitude of 1100 m), where the nutrient (OM,

CaCO<sub>3</sub>, P, K, Fe, Zn, Cu) content of the soil was higher. Generally, it can be concluded that in addition to altitude, the soil influences the essential oils content and composition of *S. hydrangea* 

The previously reported S. hydrangea oil from Fars-Abadeh, Iran was found to be rich in caryophyllene (25.2%), 1,8-cineole (15.2%) and caryophyllene oxide (11.1%) [18]. In the oil from Esfahan, Iran the major constituent were 1,8cineole (28.4%), caryophyllene oxide (16.4%), pinene (14.6%) and -pinene (14.2%) [37], whereas in the oil from Dehaghan the main constituents were -caryophyllene (33.4%) and caryophyllene oxide (25.4%) [16]. Kotan *et al.* [17] identified 54 components in the oil of S. hydrangea from Turky. Camphor (54.2%), -humulene (4.0%),cis-sesquisabinene hydrate (2.8%), myrtenol (2.6%), -bisabolol (2.2%) and 1,8cineole (2.1%) were found to be predominant components. According to the results of GC-MS analysis of S. hydrangea oil from Fars-Tekab, Iran, -caryophyllene (26.2%), 1,8-cineole (14.2%) and -pinene (11.2%) represented the major compound [18].

This prompts speculation as to the role of not only the genetic composition of the plants and geographic variation but also the nutrient status of the substrate in determining the quantity and quality of *S. hydrangea* oil.

Several authors considered the type and composition of the soil as one of the determinant factors in secondary metabolites composition and that of volatiles in particular, in addition to other explanations for the differences found in oils of the same species [30]. Pluhár et al. [7], through assessment of environmental factors affecting the essential oil of T. pannonicus and T. praecox found that quantity and quality of the essential oil existed in T. pannonicus depends on pH, K CaCO<sub>3</sub>, Mn, Mg, Fe, Co and Cr in the soil while, in T. Praecox, high levels of thymol depends on the concentrations of Na, K, Mn and Cd in the soil. Habibi et al. [31] expressed that there was different correlation between the elements existed in the soil and the quality of the essential oil of T. kotschyanus. In addition, Figueiredo et al. [30] working on the specie Achillea plarmica concluded that there was a significant relationship between the amount of the essential oil compounds and the soil and bioclimatic factors.

Although different soil properties affect the growth and the increase in the quantity and quality of the substance in medicinal plants, we cannot only rely on the soil properties. The different essential oil compositions of a species found for different origins reflect the different environmental conditions of each particular location (different altitudes, different solar exposition, different soil types, etc.). We cannot forget that all these things mix together, so the differences in the oil composition found for different geographical origins are also due to genetic differences.

#### Conclusion

In our study, it has been shown that the oils of wild growing S. hvdrangea in Kiarar-Hezarjarib, Iran is rich in 1,8-cineole, camphor, -pinen, naphthalene, bicycleheptan, -cadinene, and ylangene. In lower altitude, 1,8-cineole and camphor were the major components respectively while, in higher altitude naphthalene and 1,8-cineole were dominant components respectively. This study showed that the altitude and soil properties play a significant role in the oil yield and constituents in S. hydrangea However, the higher essential oil efficiency and compositions were found in the higher altitude, percentage of the essential oil compounds was more in the lower altitude where the nutrient content of the soil was higher. We can conclude that at lower altitudes there are more optimal conditions for the plant harvest and taking essential oil. Moreover, the occurrence of different oils of a species from different geographical areas may also be determined by the processing of the material after harvest.

Knowledge of the compositions of the indigenous medicinal plants in Iran can assist in employing the medicinal plant resources. Results obtained from investigating the essentials oils can assist in standardizing the medicinal products. In addition, considering the effect of various environmental factors, further studies are recommended to ascertain *S. hydrangea* chemical profile because of the geographical variations in Iran, and the vast distribution of *S. hydrangea* in Iran.

## Acknowledgements

The work was conducted as part of scientific research. We acknowledge the scholarship grant (grant number P.8.16.394) from the University of Zabol, Department of Range and Watershed Management. The assistance of the laboratory of the Agricultural and Natural Resources Research

Center, Mazandaran, Iran for analysis of essential oil composition is gratefully acknowledged.

## References

- Bayrak A, Akgul A. Composition of essential oils from Turkish Salvia Species. Phytochemistry.1987;26:846-847.
- Jenks AA, Kim SC. Medicinal plant complexes of *Salvia* subgenus Calosphace: an ethnobotanical study of new world sages. J. Ethnopharmacol. 2013;146:214-224
- Rajabi Z, Ebrahimi M, Farajpoura M, Mirza M, Ramshini H. Compositions and yield variation of essential oils among and within nine *Salvia* species from various areas of Iran. Ind. Crops Prod. 2014;61:233-239.
- 4. Rechinger KH, Hedge IC. Flora Iranica Akademiche Druck verlag sanstalt, Gras, Austria. 1982
- 5. Mahdavi M, Jouri MH, Mahmoudi J, Rezazadeh F, Mahzooni-Kachapi SS. Investigating the altitude effect on the quantity and quality of the essential oil in *Tanacetum polycephalum* Sch.-Bip. polycephalum in the Baladeh region of Nour, Iran. Chinese J Nat Med. 2013;11: 553-559.
- Omidbeigi R. Production and manufacturing the herbs. Mashhad, Beh-nashr Publication, Iran. 2005.(In Persian).
- Pluhár Z, Héthelyi E, Kutta G, Kamondy L. Evaluation of environmental factors influencing essential oil quality of *Thymus pannonicus* All and *Thymus praecox* opiz. J Herbs, Spices Med Plants. 2007;13:23-43.
- 8. Abbaszadeh B, Sharifi Ashorabadi E, Lebaschi MH, Paknejad F, Ardakani MR. Effect of application methods of nitrogen fertilizer on essential oil content and composition of balm (*Melissa officinalis* L.) under field condition. Iranian J Med Arom Plants. 2006;23:183-193. (In Persian).
- Ardakani MR, Mafakheri S. Designing a sustainable agroecosystem for wheat (*Triticum aestivum* L.) production. J Appl Environ Bio Sci. 2011;1:401-413.
- Jenks AA, Kim SC. Medicinal plant complexes of Salvia subgenus Calosphace: an ethnobotanical study of new world sages. J. Ethnopharmacol. 2013;146:214-224.
- Alonso-Amelot ME, Oliveros-Bastidas A, Calcagno-Pisarelli MP. Phenolics and condensed tannins of high altitude *Pteridium arachnoideum* in relation to sunlight exposure, elevation, and rain regime. Biochem. Syst. Ecol. 2007;35:1-10.
- 12. Pérez-Estrada LB, Cano-Santana Z, Oyama K. Variation in leaf trichomes of Wigandia urens: environmental factors and physiological consequences. Tree Physiol. 2000;20:629-632.
- 13. Lommen WJM, Bouwmeester HJ, Schenk E, Verstappen FWA, Elzinga S, Struik PC. Modelling processes determining and limiting the production of secondary metabolites during crop growth: the example of antimalarial artemisinin produced in *Artemisia annua*. Acta Hortic. 2008;765:87-94.

- Ncube B, Finnie JF, Van Staden J. Quality from the field: The impact of environmental factors as quality determinants in medicinal plants. S. Afr. J. Bot. 2012; 82:11-20.
- Jaakola L, Maatta-Riihinen K, Karenlampi S, Hohtola A. Activation of flavonoid biosynthesis by solar radiation in bilberry (*Vaccinium myrtillus* L.) leaves. Planta. 2004; 218:721-728.
- 15. Rustaiyan A, Masoudi SH, Jassbi A. The Composition of the essential oil of *Salvia hydrangea* DC.exBenth. J. Essent. Oil Res. 1997; 9: 599-600.
- 16. Barazandeh MM. Volatile constituents of the oil of *Salvia hydrangea* from Iran. J. Essent. Oil Res. 2004;16 :20-21.
- Kotan R, Kordali S, Cakir A, Kesdek M, Kaya Y, Kilic H. Antimicrobial and insecticidal activities of essential oil isolated from Turkish *Salvia hydrangea*. Biochem. Syst. Ecol. 2008;36:360-368.
- Sonboli A, Kanani M, Yousefzadi M, Mojarad M. Chemical composition and antibacterial activity of the essential oil of *Salvia hydrangea* from two localities of Iran. Iranian J Med Plants. 2009;2:20-28. (In Persian).
- Wang A, Luo C, Yang R, Chen Y, Shen Z, Li X. Metal leaching along soil profiles after the EDDS application– A field study. Environ. Pollut. 2012; 164: 204–210.
- Thomas GW. Soil pH and soil acidity, In: Sparks DL (ed.). Methods of soil analysis, American Society of Agronomy, Soil Science Society of America, Madison, Wisconsin, 1996, pp. 475-490.
- Rhoades JD. Salinity: Electrical conductivity and total dissolved solids, In: Page AL (ed.). Methods of soil analysis. American Society of Agronomy, Madison, Wisconsin, 1996, pp. 417-435.
- Lo I, Tsang D, Yip T, Wang F, Zhang WH. Influence of injection conditions on EDDS-flushing of metalcontaminated soil. J. Hazard. Mater. 2011; 192: 667–675.
- Bray RH, Kurtz LT. Determination of total, organic and available forms of phosphorus in soils. Soil Sci. 1954; 39-45.
- 24. Knudsen D, Peterson GA, Pratt P. Lithium, sodium and potassium, In: Page AL (ed.). Methods of Soil Analysis. American Society of Agronomy, Soil Science Society of America, Madison, Wisconsin, 1982, pp. 225-246.
- 25. Wilcox LV. A method for calculating the saturation percentage from the weight of a known volume of saturated soil paste. Soil Sci. 1951;73:233-238.
- Allan JE. Atomic-absorption spectrophotometry with special reference to the determination of magnesium. Analyst. 1958;83:466-471.
- Allison LE, Moodie CD. Carbonates, In: Black, C.A. (ed.). Methods of Soil Analysis. American Society of Agronomy, Soil Science Society of America, Madison, Wisconsin, 1965, pp. 1379-1400.
- Adams RP. Identification of essential oil components by gas chromatography/mass spectrometry, Allured Press, Carol Stream, IL. USA, 1995.

- Coley PD. Interspecific variation in plant anti-herbivore properties: the role of habitat quality and rate of disturbance. New Phytol. 1987;106:251-263.
- Figueiredo AC, Barroso JG, Pedro LG, Scheffer JJC. Factors effecting secondary metabolite production in plant: volatile. components and essential oils. Flavour Fragr. J. 2008; 23:213-226.
- 31. Habibi H, Mazaheri D, Majnoon-Hassani N, Chaeechi MR, Fakhr-Tabatabaee M, Bigdeli M. Investigating the effects of altitudes on the essential oil and herbal compositions of *Thymus kotschyanus* in Taleghan. J Res Construct. 2006;19:2-10.
- 32. Haider F, Kumar N, Banerjee S, Naqvi AA, Bagchi GD. Effect of altitude on the essential oil constituents of *Artemisia roxburghiana* Besser var. *purpurascens* (Jacq.) Hook. J. Essent. Oil Res. 2009;21: 302-303.
- 33. Mahzenoozi-Kachpi SS, Mahdavi M, Jouri MH, Akbarzadeh L. The effects of altitude on chemical compostions and fuction of essential oils in *Stachys lavadulifolia* Vahl. (Iran). International J Med Arom Plants. 2014;4:107-116.
- 34. Senatore F, De Fusco R, De Feo V. Essential oils from *Salvia* spp. (Lamiaceae). I. Chemical composition of the essential oils from *Salvia glutinosa* L. growing wild in southern Italy. J Essent. Oil Res. 1997;9:151-157.
- 35. Mirjalili M, Fakhr Tabatabaie M, Omidbeigi R. Study of compatibility and evaluation of the essential function in the indigenous biomass of the Iranian lemongrass. Iranian J Agric Sci. 2006;36:33-41. (In Persian).
- 36. Dehghan Z, Sefidkon F, Bakhshi khaniki GH. Effects of some ecological factors on essential oil content and composition of (*Ziziphora clinopodioides*. Suamb sp. *rigida* Boiss.). Iranian J Med Arom Plants. 2010;26:49-63. (In Persian).
- 37. Ghannadi A, Samsam-Shariat H, Moattar, F. Composition of the leaf oil of *Salvia hydrangea* DC.exBenth. grown in Iran preview access options. J Essent Oil Res. 1999;11:745-746.