



Investigating the Effects of Major Ecological Factors on Essential Oils and Existing Chemical Compounds of *Nepeta menthoides* in Habitat Conditions

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ABSTRACT

A native Iranian plant, *Nepeta menthoides*, located and distributed in the north west of Iran, is a gramineous, perennial plant species which belongs to mint family. Highlands of Sabalan Mountain, in Ardebil province, Shabil and Gotur Suyi regions are habitats for this specific species. This study enjoys samples collected from selective habitats in the stage of 50% flowering; moreover, the collected samples were dried in an appropriate and standard condition. Abooreyhan laboratory of Jungle and Pasture research institute was the place in which the above mentioned samples have undergone the process of essential oils, using the water steam method of distillation. The extracted components were gauged and identified by means of gas chromatography system connected to mass spectrograph. The results of the study distinctly indicated that in the habitat condition, the species grows in the highlands of Sabalan Mountain over the attitude of 3200 to 4000 meters, this specific species is highly visible in series and sandy clay hills, it is demonstrated that the density of the species has been increased as the height was increasing. Essential oil in the investigated area was ranging from 1.6% to 2.40%, admittedly, the least amount was in Gotur Suyi, while, the highest amount belonged to Shabil. Nineteen chemical compounds were identified in samples collected from natural habitats, amongst all, the highest proportion belongs to 8,10-Cineole and Citronellol acetate. 8,10-Cineole was ranging from 33 to 34 percent, the least amount was identified in Gotur Suyi region (before hot water spa) and the highest amount was observed between Gotur Suyi and Shabil. As mentioned earlier, Citronellol acetate was the second chemical compound, which was ranging from 16% to 31% and the least and the highest amount were known in Gotur suyi and Shabil, respectively. Samian medicinal plants research station possesses the highest level with the value of 57%.

Key words: Performance, Chemical compounds, *Nepeta menthoides* (collected from Sabalan)

Introduction

The use of the chemical medicines to cure and treat some diseases caused a complicated problem called, medicines side effects. Some side effects and negative consequences culminated in limiting the use of some medicines. Due to the fact that medicinal plants are more compatible with the nature, these are considered to be a precious source of medicines of which human beings are in need. Some active and common compounds extracted from plants are usually used in various parts as food additives, pigment, pesticides and air freshener, etc. These compounds, made in organs and tissues of medicinal plants, are totally recognized as secondary metabolites (Tesay, & Nasiri, 2004). Considering climatic, geographical situations and potential capability of medicinal plants growth, Iran is known as one of the best places in the world. This country, in the past, was a source of production and consumption of medicinal plants. Beside the ever-increasing importance of medicinal plants, which are rapidly replacing the chemical drugs in the

world, exporting these plants can be assumed a good economic source for the country. Unfortunately, though, there are rich sources of these plants in Iran, there is no enough attention and awareness allocated to this source, most of these plants are left untouched and unused (Shariyat, 2003). Ardebil province enjoys a sufficient variety of the plants mainly because of having climatic variety (moderate semi steppe, in Moghan and bank of Ghezel Ozan, cold semi steppe, ultra cold semi steppe in mountainous highlands) and altitude difference from the sea level which is between 20 meters from bank of Aras river and Moghan region, and 4811 meters from mount Sabalan. Because of all mentioned factors, local medicinal plants in the province are so numerous that each has its own abilities and features. *Nepeta menthoides* located and distributed in the north west of Iran is a gramineous, perennial plant species which belongs to laminaceae family, Shabil and Gotur Suyi regions are habitats for this specific species (Azimi, 2000). Despite of being aromatic, this kind of Sabalan pennyroyal is less likely to be used by local people, and it is known as "Yava Yarpooz" (literally translated as "bad pennyroyal"). It has some chemical compounds similar to other pennyroyal which seems to have

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unique and noticeable medicinal properties (Sajjadi, 2005). Having described all, the aim of the study is to investigate the effects of different highlands, essence level and percentage of chemical compounds of pennyroyal in Sabalan and investigate the effect of different domain, essence level and percentage of chemical compounds of pennyroyal in Sabalan.

Materials and Methods

In order to conduct this study, at first, library studies including surveying new references such as flora, articles and results of researches regarding local medicinal essence plants and some consultation with professors and experts in Ardebil and its habitats are carried out. Then, field operation was started to find natural habitats of mentioned species made to determine the locations and essential note taking in terms of slope, altitude and, soil, etc. Then, some samples were collected from selective habitats in the stage of 50% flowering. Collected samples were dried in proper and standard places. Abooreyhan laboratory of Jungle and Pasture research institute was the place in which the above mentioned samples were used to be extracted with resorting to the water steam method of distillation. Gas chromatography system and gas chromatography connected to mass spectrograph were utilized to measure and identify the extracted components. Features of above mentioned system are following. Specifications of gas chromatograph and gas chromatograph connected to mass spectrograph:

Gas chromatography (GC): Model GC-9-A Shimadzu utilized with FID Detector (Ionization by hydrogen flame) and chromatopae C-R3A data processor. A DB-1 Column which is a non-polar one with the length of 60 meters and inner diameter of 25.0 mm and its thickness of the stationary phase accounts for 25.0 μm . Carrying a gas called Helium with pressure of 5.2 kg/cm^2 at the very beginning of column. Gap ratio of 1:100 was used to dilute the sample. The injection temperature of 220 °C and 250 °C for detector temperature were detected.

Chromatography connected to mass spectrograph(GC/MS): Gas chromatography system (model: Varian) is connected to mass spectrograph (model: Saturn II). A DB-1 Column with the length of 60 meters and diameter of 250 μm and its thickness of the stationary phase accounts for 25.0 μm , head gas field pressure 35psi. Ionization energy accounts for 70ev. Thermal planning field, temperature ranging

from 40 to 250 °C with the speed increase of 4°C per minute and the temperature was 260 degrees injection chamber.

Results and discussion

The result of the current research have clearly showed that the species, in the habitat condition, grows in the highlands of Sabalan mountain over the height of 3200 to 4000 meters, they are highly visible in series and sandy, clay hills. It has been proved that as we were going to higher areas, the density of this species has been increased. The essential oil in the investigated area was ranging from 1.6% to 2.40%, additionally; Gotur Suyi had the least amount, whereas the highest amount belonged to Shabil. 19 chemical compounds were recognized in samples from natural habitats, (table 1) among all, 8, 10- Cineole and Citronellol acetate possessed the highest proportion. 8, 10- Cineole was ranging from 33 to 34 percent, the least amount was determined in Gotur Suyi region (before hot water spa) and the highest amount was seen between Gotur Suyi and Shabil. As discussed earlier, the second chemical compound, Citronellol acetate, was ranging from 16% to 31% and the least and the highest amount were known in Gotur Suyi and Shabil, respectively. The highest level of this compound with the value of 57% belongs to Samian medicinal plants research station. A study, conducted by Kiomarsi (2013), indicated that Sabalan pennyroyal regarding respiratory organs' performance has the highest value in Ardebil, and Meshkin Shahr, moreover, if one can consider the essential oil, Khalkhal and Samian station have the highest standard. The main reason which causes low respiratory organs' performance in Khalkhal station can be related to climatic and soil condition dominant there. Difficult condition as proved on other studies increases the essential oil. Studies (Akbar Esmaeili & Hamzeh Amiri, 2008) revealed that chemical compounds in Khalkhal, Ardebil, and Meshkin Shahr are 45, 39, and 37%, respectively. The highest chemical compound with 39.9% is related to 1, 8-Cineole in Samian station. *Teucrium polium L.* was analyzed by GC/MS and GC system. 44 compounds were detected in its essential oil that accounts for 91.76% of whole essence. The main components of the essential oil in this species are the followings: Alpha-pinene (13.95), beta-caryophyllene (12.35%), Germacrene B (11.74%), Betapynene (8.75%), and limonene



(7.60%) (Esmaili & Amiri, 2008). Ghanj Ali & Pour Ramazani Harati (2012) investigated the anti-bacterial and anti-fungal effects of essential oils in Kermani Artemisia plant. Kermani Artemisia plants were collected from almost about 1800 to 2000 meters highlands in Fariyab region in 2011. They extracted the essential oils of this plant using the water method of

distillation with the efficiency of 1.76%. The extracted essence was analyzed by GC/MS system and 41 components were detected that main ones are as followings: 1, 8 Cineol (26.93%), Camphor (16.97%), α -Thujen (7.52%), Borneol (7.47), α -Terpineol (5.77%) (Ghanj Ali & Pour Ramazani Harati, 2012).

Table 1 .Chemical composition of Nepeta menthoides in 6 points of natural habitat

PK.NO	PK	1	2	3	4	5	6
1	<i>Alfa - thujene</i>	0.298	1.48	0.80	1.14	0.743	0.841
2	<i>Alfa – pinene</i>	1.196	1.96	2.30	2.04	1.355	2.118
3	<i>Sabinene</i>	2.073	2.81	3.28	3.311	3.12	3.38
4	<i>Beta – pinene</i>	5.12	5.63	8.84	6.15	5.28	6.96
5	<i>Myrcene</i>	0.55	0.77	0.9997	1.199	0.62	1.150
6	<i>P – cymene</i>	0.825	0.33	1.21	-	0.305	0.68
7	<i>1,8 – cineole</i>	33.27	30.96	57.33	38.46	41.920	50.094
8	<i>Gama - terpinene</i>	0.31	0.187	0.30	0.331	0.0523	0.430
9	<i>Cis sabinene hydrate</i>	0.380	1.63	0.036	2.52	1.530	2.24
10	<i>Linalool</i>	0.1697	0.414	1.503	0.670	0.154	0.441
11	<i>Isopentyl isovalerete</i>	0.43	0.327	1.384	0.602	0.010	0.453
12	<i>Undecanc</i>	0.41	0.231	1.131	0.28	0.068	0.459
13	<i>Sigma - terpineole</i>	1.25	1.127	2.36	1.23	1.31	1.16
14	<i>Terpinen – 4 – ol</i>	2.313	1.16	2.89	0.66	1.31	1.06
15	<i>Alfa - terpineol</i>	3.53	2.41	3.87	3.43	3.172	4.05
16	<i>Citronellyl acetate</i>	16.143	36.075	2.03	12.21	17.53	0.422
17	<i>Geranyl acetate</i>	4.90	6.59	8.15	6.33	15.41	4.65
18	<i>Beta – bisabolene</i>	1.23	2.24	0.61	3.71	0.93	3.05
19	<i>Cis nerolidol</i>	23.67	3.68	0.059	1.675	5.143	2.93
20	<i>B-himachalene</i>	1.51	-	-	-	-	-

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