

Ethno Pharmacology and the Investigation of the Most Important Secondary Materials and the Comparison of Chemical Combinations of Essential Oil of Different Organs

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Abstract: *Perovskia abrotanoides* Karel. is one of the valuable medicinal species in the North of Iran and it is growing wild in the margin of mountainous roads of arid and cold climate of Golestan and Khorasan Shomali. It is for a long time that indigenous people by different methods in traditional medicine use its products in preventing and curing diseases. In this research beside obtaining ethnopharmacological data to investigate and compare the most important secondary active materials of different organs the soil with the texture of clay loam, gravel loam and neutral pH collected from two natural habitats located in Golestan (Shahkuh 2300m) and Khorasan-e Shomali (Chaman Bid 1076 m). Extracting for the analysis of active combinations of organs are made in habitat and it was done by Pearson chi square and it was significant at $P < 0.05$. The results showed that the plant growth is starting from early May to early September and flowering is done in July as in the mountain in addition that the color of the plant is increased and phonological cycle is done with delay. The results of ethnopharmacological investigations of local shepherds and healers in both regions shows the ecological importance of that species and this plant is used in grazing and traditional medicine with other medicinal and local plants in the region. This plant is mostly used as fortifier, antiseptic, rheumatic pains, anti-inflammatory, leishmaniasis, anthelmintic. In this research the results of the investigation of the most important plant secondary materials in both regions showed that effective essential oil and also flavonoid, total phenolic, anthocyanin and antioxidants are different in quality and quantity in different organs and different habitats and by the increasing height of habitat, they are increased namely in floral aerial that is probably in proving the belief of indigenous people based on their interest in collecting and consuming this plant in mountainous area of Shahkuh. The sample was extracted by distilled water. The output of essential oil in Chaman Bid (Khorasan Shomali) in the leave is 2.53%, floral areal 0.81% and stem is without essential. But in Shahkuh (Golestan), the essential output in the stem is 0.34%, in the leave 1.42% and in floral aerial 1.58%. Then the samples were analyzed by Gas chromatography (GC) and Gas chromatography-mass spectrometry (GC-MS). The significant combinations in organs are terpinolene (0-24.7%), Verbenone (4.9-31.2%), sabinene (1.7-16.8%), terpinen-4-ol (0-43.8%) and γ -terpinen (0.9-19.8%).

Key words: *Perovskia abrotanoides* Karel., Ethnopharmacology • Essential chemical combinations • Different organs • Secondary effective materials • Iran (natural habitats of Golestan and Khorasan-e Shomali)

INTRODUCTION

Considering the economical value, especial position of aromatic medicinal plants in community health, they are considered more by scientific and research centers. Global trend is toward applied researches about the identification of the species, habitat conditions, obtaining important ethnopharmacological data from local

healers and above all the investigation of the effective secondary and medicinal material with the aim of formulation and production of effective and safe natural medicines in accordance with their performance in traditional medicine [1].

It is estimated that today 70 percent of the communities in the world use various medicinal plants in different forms in daily life; diet and disease cure [2].

Scientists view about the role of medicinal plants in the economical development is changed its direction despite all changes and the lack of considering it is shifted to its development, even in industrialization by permanent development. Thus, the importance and role of research in this sector is obvious for all.

Summer and arid rangelands of Golestan and Khorasan Shomali in Iran is a gential storage and valuable treasury of aromatic medicinal plants with unique properties. Although these species are used for a long time in traditional medicine, due to unknown nature of these medicines, unduly grazing and above all the lack of doing environmental studies are not recognized and they are decreasing gradually. *Perovskia* L. is the local name of *Perovskia abrotanoides* Karel. belonging to the family of Lamiaceae in Iran and is consisting of 3 species and *P.abrotanoides* species is extensively found in these two provinces [3] and are growing wild in Iran, Afghanistan, Pakistan and Turkmenistan and is found as an aromatic plant in Golestan, Isfahan, Khorasan, Mazandran and Sistan and Baluchistan [4-7].

In Pakistan *P.abrotanoides* is used as refrigerant [8]. In china, *P.abrotanoides* is used in curing atherosclerosis, cardiovascular diseases, liver fibrosis and other diseases [9]. The studies of traditional medicine showed that *P.abrotanoides* is used as a pain-killer and anti-inflammatory and this is proved in animal model researches [6, 9, 10].

In another research extracts of *P.abrotanoides* showed that the herbal tea of this plant is used in curing infection problems and painful urination two times a day for 8-10 days [11].

Rustaian *et al* [12] indicated that this plant is used as refrigerant. Also the anti-bacterial activities of its essential oil is reported [12]. Its antioxidant performance including heart enhancing, inhibiting eduz Reductekaz and its optimized performance as cell toxicity in pathogens, viruses and cancer cells are reported [8, 9]. Researches showed that essential of *Cuminum cyminum* and *Perovskia abrotanoides* Karel. is consisting of anti-pathogen against storage pest and they are the most important chemical combinations of essential of monoterpenoids that are mostly used as pesticides [7]. In another research the anti-plasmodium property of floral aerial of *P.abrotanoides* against *Plasmodium falciparum* parasite [13]. In the south of Iran, the dried stem powder of this plant is used for treating dermal leishmaniasis and its medicine is used on wound to relieve pain [5]. So considering the scatter and natural habitats of different species of *Perovskia abrotanoides* Karel. and

also medicinal consumption of this plant in traditional medicine of the province as single or in combinational with other plants, in this research we identified the most abundant habitats of *Perovskia abrotanoides* Karel. in two provinces (*P.abrotanoides*), phenology, ethnopharmacology, the comparison of the most important secondary combination and also the studied species by hydrodistilling in a Clevenger type was extracted and the combinations were identified by GC and GC/MC in two different habitats of Golestan and Khorasan.

MATERIAL AND METHODS

Collection and Identification: Field research was done for ecological, phonology, identification and plant crop investigations at least for two natural habitats of the studied species (1000m in Chaman Bid of Khorasan Shomali province and 2300 m in Golestan province Shahkuh) during a one year period (2008-2009). In each region, 20 bases of the plant that were morphologically the same, were selected and marked and each month they were visited consecutively and the occurrence date of vital phenomena of the plant was recorded to drying stage. Also, in field research, important traditional data was obtained about the optimized place and time with the aim of collecting edible organ of *Perovskia abrotanoides* Karel. and its consuming method was asked by local shepherds and experienced people. The collected samples of the plant was identified and it was prepared for extraction in its dry powder lab.

Extracting: Ground, dried leaves of the plant (500 mg) were extracted, with 10cc of methanol for 10m then, it was filtered, again 10cc methanol was added and again it is filtered. Then we pour filtered solutions on each other and then dry it in water bath of 80°C (the liquid is evaporated completely), finally we add 5cc of methanol that is stuck to the bottom of the dish, then we shake it to be solved.

Flavonoid Assay [14]: To 0.5cc of methanol extract we add 0.1cc Aluminum chloride $AlCl_3$ 10% in methanol, then 0.1cc potassium acetate of 1 molar and 2.8cc distilled water is added. After 30m being kept at room temperature, its absorbencies is measured at 415nm. The standard curve is calculated based on Gallic acid and the amount of Flavonoid quercetin mg/gr was determined.

$$A=0.0067C+0.0132$$

Assay for Total Phenolic [15]: To 1cc of methanol extract, we add HcL 1cc (6M) and 5cc methanol of 75% and pour it in tubes and keep it in water bath of 90°C for 2 H and it is shaken during this time and they are cooled at room temperature for a while, then they reached to the volume of 10ml by distilled water, then, we take 1cc of the above solution and add 5cc pholine (1:10) and 15ml $(\frac{7gr}{100})Na_2CO_3$ and then reach the volume of 100ml and its absorbencies is measured at 760 nm (Blank: Methanol).

Standard curve is calculated based on Gallic acid and the amount of Total phenolic equivalent to Gallic acid was measured in each milligram dry powder.

$$C=0.229A + 0.00217$$

Total anthocyanin assay [16]: We pour 0.4 cc methanol extract of the plant in two separate tube, we add 3.6 ml buffer of potassium chloride to one of the tubes (0.025M) at PH=1 and we add 3.6 ml of sodium acetate (0.4M) at PH=4.5 and absorbencies of each tubes are measured at two wave length 700 nm, 510nm. (Blank: Distilled water) Based on milligram, anthocyanin is measured equivalent to cyanidin-3-glucosid per gram.

$$A = (A_{510} - A_{700})_{PH1} - (A_{510} - A_{700})_{PH4/5}$$

$$TAC = \left(\frac{A \times MW \times DF \times 100}{MA} \right)$$

A: absorbencies

MW: 499.2

Antioxidant assay by Tris-Hcl [17]

At First Extracting Is Done by the Following Method: To 1gr powder of the plant, 10ml solution (of ascertic acid H₂O (40) (21) Ethanol (60)) is added, then after 1 hour it is filtered and again 10ml of the solution is added and then after half an hour it is filtered and reach 25ml volume with the above solution.

Antioxidant Assay by Tris-Hcl

Buffer: 12.2gr Tris is solved in 400 ml water and set PH with HCL:6m ON 7.4 and reach the solution to 1000 ml. 19.7 mg DPPH is reached to the volume of 100 by ethanol. **Sample:** 0.5 ml extract and 4ml buffer of Tris and 5 ml DPPH is poured in volumetric flask and is reached to the volume of 10 by buffer and then it is poured into test tube and put it in tube case.

Sample Blank: 1 ml of the extract and 9 ml of the buffer is poured into volumetric flask and reach to the volume of 10 and then 10ml of ethanol 0.96 is added.

Control: 0.5 ml water and 2 ml, DPPH and 4.5 ml buffer are poured into volumetric flask and reach the volume of 10 by ethanol.

Blank: 1ml water and 9 ml buffer is poured into volumetric flask to reach volume of 10 by buffer, then 10 ml ethanol is added. After 30 m we read absorbencies at the wavelength of 517nm. Control absorbencies is at first multiplied by 2.5 and then we will have the following formula:

$$\text{Control sample } AAc = \left(1 - \frac{A}{A} \right) * 100$$

Extract Method: Dry samples of the plant were extracted by hydrodistilling method for 2 h and essential output of Chaman Bid region of Khorasan Shomali in the leaf is 2.53% and in floral aerial 0.81%, while it didn't have essential at stem and in Shahkuh of Golestan province, essential output at the stem was 0.34%, in the leaf 1.42% and in floral aerial 1.58%.

Gas Chromatography (GC): Gas Chromatograph (Thermo-UFM) equipped with flame ionization detector (F.I.D) was carried out using a data processor with the software of Chrom-card 2006, equipped with ph-5 columns (10m × 0.1mm; film thickness 0.4 micron). Column temperature was held at 60°C to 285°C with the speed of 3°C per min is done at 5.8min. With helium as carrier gas and its pressure at the beginning of the column is 3kg/cm², split ration as 1:100, for diluting the sample, the temperature of the injector is 280°C and detector temperature was held as 280°C.

Gas Chromatography and Mass Spectrometry (GC/MS): Gas chromatography (Varian 3400) coupled with mass spectrometry by software Saturn II, the column is similar to GC device. Pressure of the top of the column is 35Psi, ionization energy 70 electron volt. Thermal programming of the column is ranging from 40 to 250°C with the rate of 4°C per min, injection room temperature 260°C and the temperature of transfer line is 270°C.

Table 1: Chemical compositions of *Perovskia abrotanoides* Karel. essential

No.	Compositions	Floral aerial of Golestan (Shahkuh)	Floral aerial of chaman bid (Khorasan Shomali)	Leaf of Golestan (Shahkuh)	Leaf of chaman bid (Khorasan Shomali)	Stem of Golestan (Shahkuh)
1	sabinen	12.2	16.8	4.6	2.8	1.7
2	myrcene	3.5	6.3	4	6.4	1.5
3	α -phellanderene	4	5.7	4.3	----	1.7
4	p-cymene	1.1	1.6	0.6	----	0.5
5	(Z)- β -ocimene	6.8	1.3	9.5	----	2.3
6	γ -terpinene	0.8	0.9	0.8	19.8	----
7	terpinolene	22.6	24.7	22.5	----	10
8	verbenone	25.1	31.2	29.9	4.9	25.6
9	trans-carveol	5.2	2.2	4.9	2.6	3.2
10	isobutyl benzoate	½	0.8	2.1	0.6	4.7
11	α -copaene	1.7	0.3	1.7	----	3.3
12	α -himachalene----	----	0.3	----	0.8	
13	β -selinene	0.7	----	0.6	1.4	1.5
14	viridiflorene	5.2	1.7	5.4	----	9.6
15	α -cadinene	3.8	1.1	3.9	0.9	7.8
16	himachalene epoxide	1.6	1	1.1	1.5	5.3
17	longifolol	0.3	----	0.4	1.7	1.2
18	methyl tetradecanoate-----	----	4.8	4.3		
19	Curcumenol----	----	----	----	1.1	
20	(E,Z)-farnesol----	----	----	----	1.3	
21	α -bisabolol oxide A	0.8	1	0.4	----	6.4
22	Camphene----	----	----	3.4	----	
23	terpin-4-ol	----	----	----	43.8	----
24	ar-turmerone----	----	----	2.5	----	

Identification of spectrums by their inhibition indices and by injection of normal hydrocarbons (C_7 - C_{25}) under similar conditions are calculated by essentials injection and computer program by Basic language. Also, their comparison are done by different resources and by mass spectrum of standard composition and available data in library of GC/Mc device.

RESULTS

The results of ecological investigations in this research showed that Karel. *P.abrotanoides* species is more scattered in comparison with other species in the province that are mostly growing wild in the margin of semi-arid mountainous and cold and semi-arid climate of Golestan (2300m) and Khorasan Shomali (1000 m) and in soils with the neutral PH of to alkali, Ec(0.53-2.5) in the light texture of gravel loam and clay loam.

From phonological results, from early May to early September this plant is growing. Flowering period of this plant is from July to September. By the increase of the height of this plant in 2300 m, in addition to the increased of effective materials, annual growth cycle of the plant is done with 20 days delay and the color is increased.

The results of ethnopharmacological investigations showed that in local culture root of Karel. *P.abrotanoides* is used to treat dermal leishmaniasis and also it is used as fortifier, antiseptics, refrigerant and rheumatic pains, anti-inflammatory, anthelmintic and anti-bacterial as single or combinational with other medicinal species of the region (*Artemisia* and *Jalas Thymus carmanicus*). Leaves of *P.abrotanoides* are used to control painful urination as herbal tea two times a day for 8-10 days.

The remaining results of this research about the investigation and comparisons of secondary active compositions of Flavonoid, phenolic, anthocyanin in floral aerial and leaf that are used mostly in traditional medicine, indicated that the amount of that compositions by increasing height, especially at 2300 m of mountainous area of Shahkuh are increased (Charts 1-4). The quality and quantity difference of the effective compositions in plant organs in two provinces are totally significant.

After the collection of samples from the leaf, floral aerial and stem, extracting essential and its analysis is started. In Chaman Bid region 21 major and minor compositions (Table 1) are identified. Major essential compositions are including terpinen-4-ol (43.8%), γ -terpinene (19.8%), in floral aerial Verbenone

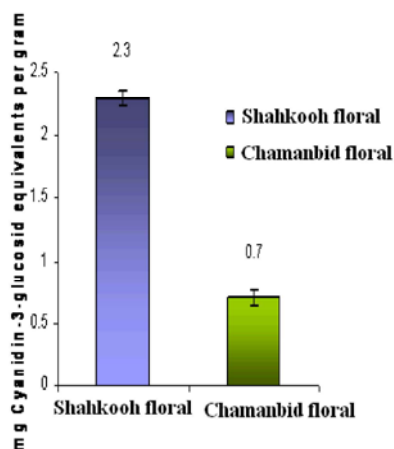


Chart 1: The comparison of the amount of anthocyanin in floral aerial in two regions.

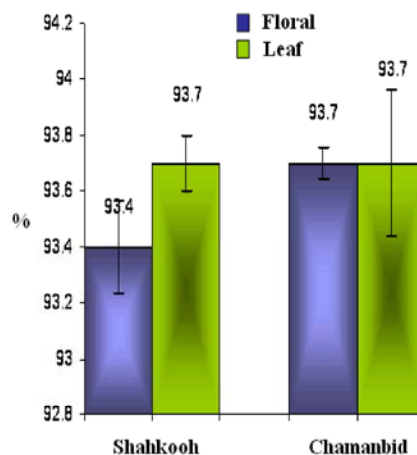


Chart 4: The comparison of the amount of antioxidants in two regions.

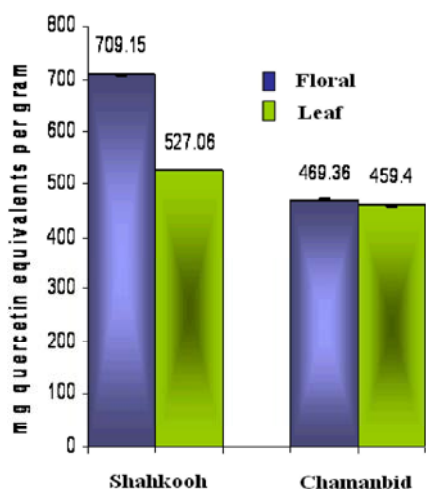


Chart 2: The comparison of the amount of Flavonoid in two regions.

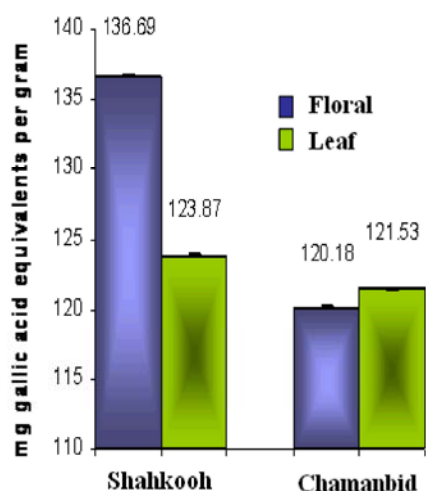


Chart 3: The comparison of the amount of phenolic in two regions.

(31.2%), terpinolene (24.7%) 'sabinene (16.8%) and there is no essential in the stem. While in Shahkuh 21 major and minor compositions (Table 1) are identified and major compositions of the essential in the stem are verbenone (25.6%) terpinolene (10%) viridiflorene (9.6%) in the leaf it is including verbenone (29.9%), terpinolene (22.5%), (Z)- β -ocimene (9.6%), in floral aerial verbenone (25.1%) terpinolene (22.6%), sabinene (12.2%), in flower verbenone (24.7%), terpinolene (19.7%), sabinene (18.7%).

DISCUSSION

Golestan and Khorasan Shomali provinces are located in the north east of Iran and are one of the natural habitats of different species of *Perovskia abrotanoides* Karel. and it is for a long time that this plant is used in the traditional medicine of the people in this region to prevent some diseases. In Pakistan *P.abrotanoides* is used as refrigerant [8]. The investigations of traditional medicine showed that *P.abrotanoides* is anti-inflammatory, anti-pain and this is proved in animal model researches [6, 9, 10].

According to the reports, villagers in the Isfahan province of Iran apply a poultice, made of crushed roots of the plant, water, sesame oil and wax, on lesions caused by cutaneous leishmaniasis [5, 9].

Leaves of *P.abrotanoides* are used to control painful urination as herbal tea two times a day for 8-10 days [11]. In Golestan province *P.abrotanoides* is used to treat leishmaniasis and dermal problems [13].

Aerial branches of *Pabrotenoides* are anti-inflammatory and anti-pain [6, 10]. In another research the anti-plasmodium property of floral aerial of *P.abrotanoides* against *Plasmodium falciparum* parasite and cytotoxic activity are shown [13].

Arabi *et al.* [7] believed that essential oil of *P.abrotanoides* plays an important role in protection of stored grains and reducing the need for it [7].

The researchers attribute the amount of essential compositions to genetical and environmental factors [18], [19]. Besides, there is a great difference between the existing combinations of essential of one species in different growing conditions [19, 20].

Anthelmintic property of this plant is due to the effective material of verbenene and terpinolene. The presence of effective material of myrcene is due to its strong antiseptic property, refrigerant, inhibiting cancer glands, anti-bacterial and strong painkiller and antioxidant. p-cymene is the main factor for anti-bacterial and anti-virus property and rheumatic pains. Anti-oxidant property, anti-tumor, anti-bacterial, gall stimulator in liver, protection of Hepatic system of this plant is due the effective material of - γ terpinene.

Jaafari *et al.*, [5] reported that ethanol extract of the stems and leaves are due to the presence of phenolic and terpinene compositions are anti-leishmaniasis and anthelmintic. This medicinal plant is proved to treat anthelmintic and leishmaniasis.

Morteza Semnani *et al.* [4] believed that due to the quality and quantity change and the difference in the performance of *Perovskia abrotanoides* essential extract, different habitat, ecological conditions change, different times for collecting the plant, drying condition and extracting and they prove the difference of effective compositions in different organs and habitats of the plant. In traditional medicine *Borago Officinalis*, *Perovskia abrotanoides* and *Citrus aurantium* as herbal tea are used for palpitation, cold, influenza, depression and stress of pregnant women.

Sajadi *et al.* [21] showed that secondary metabolite of terpinene, Flavonoid and phenolic of floral aerial of *Perovskia abrotanoides* were different in different methods of extracting and it has the important role of antioxidant, anti-inflammatory and toxicity of the cell against parasitic and bacteria nematodes [21, 22]. And it is effective in treating heart problems, Hepatitis, virus and liver cirrhosis [23].

In a similar research, Pourmortazavi *et al.* [24] reported that effective compositions of floral aerial essentials of *Perovskia abrotanoides* in Tehran are different from quality and quantity amounts of terpinene and phenolic compositions.

Kayser *et al.* [25] referred to the extract of *Perovskia abrotanoides* \times *Artemisia sieberi* \times *Berberis vulgaris* and *Pomegranate* peel as the most important factors of removing intestinal worm and leishmaniasis parasite and this proves traditional consumption of this plant with other species in treating parasite and leishmaniasis wound [26].

Obame *et al.*, [22] argued that consuming essential oil of *Perovskia abrotanoides* is effective in washing wound, anti-ring worm, dermal parasites and anti-fungus and this is due to the presence of terpinene of the essential and phenolic compositions and anthocyanin of the extract of floral aerial of the plant.

Vokovi *et al.*, [27] reported about terpinene and phenolic compositions in floral aerial of *P.abrotanoides* in vitro conditions as good anti-pathogenic property against 13 pathogens.

Mockute *et al.*, [28] reported that quality and quantity of the effective materials of essential extract of *Daucus carota*, Allelopathy function, antiseptic, anthelmintic and anti-pain in different habitats of Italy as different. In another research of flavonoid and phenolic compositions in floral aerial of the plant they were recognized as strong antioxidants and anti-inflammatory with pepper mint, ginger, turmeric for treating arthritis, rheumatism and anti-pain is doing as Diclofenac.

In this research terpinene-4-l in the extract of aerial plant of *P.abrotanoides* is used as a strong pain-killer, anti-inflammatory that it has 35% muscle loosening and anti-pain [29, 30].

Cetin *et al.*, [31] refers to phenolic composition of rosmarinic acid and terpinene-4-l in Rosemary aerial and *P.abrotanoides* as the most important anti-inflammatory compositions, antioxidants with the effect of anti-spasmodic and anti-pain in treating rheumatic, arthritis and muscle spasm.

Indigenous people of the mountain use this plant with *Artemisia*, wild rosemary and *Artemisia annua* for treating rheumatic pains and dermal inflammations.

Khaliq *et al.*, [32] in a research showed that Flavonoid and phenolic compositions and anthocyanin in floral aerial of *P.abrotanoides*, rosemary and orange peel due to the high amount of effective materials and also hesperidin, abrotanone and rosmarinic acid are used as strong antioxidant, anti-inflammatory and anti-pain.

Esmaili *et al.* [13] reported that extract of floral aerial of *P.abrotanoides* due to the presence of Flavonoid and phenolic compositions and anthocyanin are anthelmintic, antioxidant and anti-inflammatory.

Cetin *et al.* [31] in the research of invitro and invivo showed that terpenes and polyphenolics and Flavonoids of floral aerial and leaves are respectively found in *Perovskia* plants, *Eucalyptus*, *Thymus vulgaris* and *Dracocephalum* are anti-inflammatory and anti-virus and are used mostly in treating dermal wounds and Herpes Simplex.

This research showed that the amount of Flavonoid and phenolic compositions respectively in floral aerals of the plant are more than in the leaf and it is increased by the height increased of Shahkuh region to (quercetin equivalents per gram mg and 709.15 mg Gallic acid equivalents per gram 136.69) in floral aerial and (mg quercetin equivalents per gram 527.06 mg Gallic acid equivalents per gram 123.87) are increased in leaves and this case proves the consumption of flowers and leaves of this plant in treating dermal inflammations, leishmaniasis wound and the boiled leaves and aerals are used in treating arthritis, rheumatism and Patellofemoral. It is also used to treat intestinal worm, parasite along with *Dracocephalum* and along with majorana it is used to prevent food spoilage in the mountain (namely to prevent red meat spoilage in the mountain).

Vardar *et al.*, [33] referred to thymol, gamma-terpinene as the most important antioxidants and anti-bacterial in floral aerial of *P.abrotanoides* and *Thymus pectinatus* and the quality and quantity amount of these materials are different in habitats.

Cetin *et al.*, [31] referred to phenolic compositions of the plant extract as stimulator, expectorant, anti-inflammatory, antiseptics and anti-bacterial. In this research the Flavonoid and phenolic compositions of floral aerial and leaf in 1000 m are respectively quercetin equivalents per gram mg 469.36 mg Gallic acid equivalents per gram 120.18) in floral aerial and quercetin equivalents per gram mg 459.40 mg Gallic acid equivalents per gram 121.53) in leaves. So it can be expected that the anti-bacterial and anti-pathogenic of this plant is more in Shahkuh region.

The most important chemical compositions of aerial organs essential oil in *Perovskia abrotanoides* Karel, were identified as 29 compositions (98.9% of the total composition) 1,8-Cineole (32.4%), myrcene (13%), α -pinene (10.2%), camphore (9.1%), β -Caryophyllene (7.9%), α -humulene (6.4%), camphene (5%) and α -bisabolol (2.6%) are the main compositions found in essential oil. The oil of this plant is consisting of a great volume of monoterpenes (78.9%), while in sesquiterpenes just 20% of essential oil were found [21].

Terpinene-4-l was reported as the most important compositions of essential of the leaf in Chaman Bid. Also, one of the most important compositions is ginger and turmeric and the researches showed that it is anti-inflammatory, pesticide, anti-parasite especially against leishmaniasis and this is obvious due to the high amount of this substance in the essential of the leaf of Chaman Bid to prove the effect of anti-leishmaniasis of the leaf and root of the plant in traditional medicine.

Golshani *et al.* [26] referred to Verbenone, phenolic and Flavonoid compositions of Rosemary extract as antioxidant, anti-inflammatory and antiseptic that is mostly used for reliving joint pins, rheumatism, arthritis, muscle pains and also for treating dermal infections and Lymph Gland infection and the amount of Verbenone in the plant organs of *P.abrotanoides* in both habitats are reported as variable from 4.9 to 31% and the amount of this substance in Chaman Bid floral aerial are more.

In another research Sayyah *et al.* [34] showed that effective substance of Sabinene is one of the most important compositions of *Ferula gummosa* essential that give the plant the property of strong antiseptics, anthelmintic, anti-pain, stomachache, Rheumatism, Migraine and wound healing. The most important compositions of essential oil of different species of *Juniperus communis* especially, *J.sabina* that give the plant the property of anti-inflammatory, antiseptics and anti-bacterial in treating Urinary tract infection (UTI).

Thus, the presence of the above compositions in floral aerial organs and leaf of the studied plant, verbenone (4.9%-31.2%), sabinene (1.7%-18.7%) and γ -terpinene (0.8%-19.8%) proves the anti-inflammatory and antiseptics and antioxidant effect of this plant.

Fukumoto *et al.* [35] showed that p-cymene ' γ -terpinene' limonene in citrus are used as monoterpene compositions of the essential to relieve pain, rheumatism and anti-inflammatory.

Mockute *et al.* [28] referred to sabinene (28.2-37.5%), terpinene-4-l (4.6-7.5%), gamma-terpinene (6.6—9.9%) in essential of *Daucus carota* as antiseptics and anthelmintic that are in different amounts in Italy.

Battaglia *et al.* [36] referred to verbenone and camphor in Rosemary and lemon extract and essential as stimulator of hair and skin, removing dermal inflammations, cold, influenza, expectorant and also anti-inflammatory in Rheumatism and Arthritis and antiseptic in UTI diseases and the above results prove the inclination of indigenous people toward traditional medicine to use floral aerial with the extract of herb venus and nettle are used as a strong anti-inflammatory, hair care and wound healing.

In the current research the most important compositions in the essential of different medicinal speices organs of *Perovskia abrotanoides* Karel in Chaman bid region obtained by hydrodistillation method in the leaf including terpinen-4-ol (43.8%), γ -terpinene (8.19%), myrcene (4.6%), verbenone (9.4%) in floral aerial, verbenone (31.2%), terpinolene (24.7%), sabinene (16.8%) and the stem is without essential. In Shahkuh reigon the major compositions identified in the essential of different organs of medicinal species *Perovskia abrotanoides* Karel that was obtained by hydrodistillaiton in stem is including verbenone (25.6%), terpinolen (10%), viridiflorene (9.6%) in the leaf verbenone (29.9%), terpinolene (22.5%), (Z)- β -ocimene (9.6%), in floral aerial verbenone (25.1%), terpinolene (22.6%), sabinene (12.2%), in the leaf verbenone (24.7%), terpinolene (19.7%), sabinene (18.7%).

Therefore, as it is shown in the results of the research, essential of one species in different regions and different period showed different compositions and this case is of great importance in the determination of the region for a good sample for industry.

CONCLUSION

Arid- cold and mountainous regions of Golestan Province and Khorasan Shomali are considered as a genetical resource and valuable treasury of various medicinal species with unique properties.

Perovskia abrotanoides Karel in the north of Iran is consisting of 3 species in which *P-abrotanoides* Karel is growing wild in the margin of mountainous roads and arid and cold climate of Golestan and Khorasan Shomali.

In traditional medicine of mountainous villages of the province, the powder of floral aerial as singular and combinational is used as fortifier, antiseptic and anti-inflammatory in treating dermal diseases, rheumatic pains, anthelmintic in combination with other medicinal and indigenous plants of the region. The results of this research showed that quality and quantity of chemical compositions of essential oil of different organs of the plant are different in two habitats and by increasing the height of the region, in addition to the change of effective substances of th essential, flavonoid, phenolic and anthocyanin compositions and antioxidants are added. The above results prove the more consumption of this plant in high land habitants due to the high amount of medicinal effective substances (flavonoid, phenolic and anthocyanin compositions and antioxidants). Besides, the similar researches in this field as discussed in this paper, prove this fact. These results prove the indigenous

people beliefs to be more inclined toward collection and consumption of the plant from Shahkuh mountainous region (2300m) of Golestan province that used dried powder by different methods in prevention and treating common diseases in the region.

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