

Examining of the quality and quantity of active ingredients of *Smirnova iranica* Sabeti in different habitats

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DOI: 10.22034/HBB.2018.11

Received: May 20, 2018; **Accepted:** June 10, 2018

ABSTRACT

In this study, four sites were chosen in the habitat of the plant which are located in Kashan. In order to complete phytochemical studies during flowering in each site, leaves were collected and after extraction of essential oils by simultaneous method of distillation and extraction using organic solvents, an analytical chromatography connected to a mass spectrometer (GC/MS) was used to identify the ingredients of essential oils. The results of the efficiency of leaves showed in different sites that the highest quantity belonged to the samples taken from sites 2 and 4. Comparison of essential oil compounds showed that the main compounds identified in all the leaf samples from all sites were compounds such as hexadecanoic acid.

Keywords: Medicinal Plant, tetradecanoic acid, Kashan, GC/MS

INTRODUCTION

Smirnovia iranica has a high economic value and very good quality due to its high protein content [1]. The vast differences between the native Iranian species and *Smirnovia turkestanica* species are leaves with three ovate leaflets, round navel of the seed and oval shape of its fruits. The results of analytical experiments of this plant showed two new isoflavonoids [2-6].

In phytochemical analysis of *Sphaerophysa salsula* in North West China that its key compounds were isoflavone, coumarin, flavonoids and sterols [7].

Evaluation of *Desmodium* (a traditional medicinal plant in China) extracts in terms of phytochemical experiments showed chemical compounds such as flavonoids, alkaloids, terpenoids, steroids, phenols, phenylpropanoid, glycosides and volatile oil [3].

The present study was carried out to determine the quality and quantity of leaf essential oil of this valuable plant in sandy fields of Kashan. It is noteworthy to mention that no research has been carried out in this field in Iran, hence this evaluation is quite unique.

MATERIALS AND METHODS

By using library studies, the habitats of the concerned species were identified. Then, considering various factors such as the availability of habitat, natural habitat conditions, the existence of different sites with appropriate distances from each other as well as the appropriate and harvestable presence of the plant through field visits from various habitats in the Isfahan province in 2012, the species were found in harvestable fields only in Rig Boland located in the North and the East of Kashan. Also, in order to compare the studied parameters in this area after repeated local visits and with regard to appropriate and harvestable presence of the concerned plant, 4 sites with a distance of 30 km from each other were selected. Geographical characteristics of the selected sites are listed in Table 1. Sampling from the studied plant leaves during the flowering in April 2012 was carried out randomly in each site in accordance with the presence and abundance of species along a 100-meter transect of the concerned plant bases.

Table 1. Geographical characteristics of the selected sites of *Smirnovia iranica* sabeti

Altitude (meters)	Latitude	Longitude	Sample	Number of site
992	N' 10.542 34°	E' 32.191 51°	1,2,3,4,5	1
1265	N' 52.505 34°	E' 50.664 51°	6,7,8,9,10	2
1451	N' 0.572 34°	E' 49.551 51°	11,12,13,14,15	3
1186	N' 0.683 34°	E' 36.910 51°	16,17,18,19,20	4

Table 2. Results of extraction of the essential oil

Essential oil Yield(%)	Weight of essential oil (g)	Site
0.03	0.02	1
0.04	0.03	2
0.03	0.02	3
0.04	0.03	4

After harvesting, the samples were transferred to the laboratory and were exposed to the open air to dry. In this research, the extraction of essential oil was carried out by simultaneous distillation-extraction method using organic solvent which was called simultaneous distillation-extraction (SDE). This method was performed based on distillation by steam and then the simultaneous extraction of essence was carried out by organic solvent from the

droplets produced by the condensation of steam.

In this regard, an amount of 40 g of leaves samples per site was transferred to a 2 liter distillation flask and then distilled water was added so that the total plant sample and distilled water could be 2/3 of the flask volume. Pentane was used as the solvent to collect the essence and this process continued for 2 h.

Immediately after extraction of essential oils, the organic solvent was removed using sodium sulfate and after evaporation of the solvent, the essence oil samples were kept in the freezer until injection into the GC-MS device.

The weight of collected essential oil was accurately calculated by an analytical scale and using the dry weight, the essential oil extraction yield was calculated.

The obtained essence samples were analyzed using GC-MS instrument containing HP-5MS column (internal diameter 0.25 mm, thickness of static layer 0.25 μm) and helium gas with 99.99 % purity. In addition, the carrier gas flow rate and ionization energy in mass spectrometer were considered as 1 ml/min and 70 eV, respectively. To identify the ingredients comprising the essential oil of the plant, the following steps were taken:

- A) According to the suggestions offered by GC-MS instrument, each of them was analyzed separately.
- B) The obtained MS spectra were compared with the spectra of reference books.
- C) With regard to the retention time of each peak, Kovats retention index (RI) of each of

them was calculated through the equation of calculating the Kovats coefficient and was compared with the Kovats index in references for similar conditions.

RESULTS

Table 2 shows the results of extraction of essential oils separately for the studied sites. Table 3 shows the types and amount of the main chemical ingredients comprising the essential oil of the leaf of the *Smirnova iranica* sabeti separately for the studied sites.

DISCUSSION

Findings about *Smirnova iranica* showed the diversity of quality and quantity of essence in the studied sites. The color of essence was pale yellow in all the sites and the highest essence yields were associated with the sites 2 and 4 with 0.04 % which represent the right conditions of these sites in terms of quantity, while studying the genus *Desmodium* (a plant of the same subcategory of the genus *Smirnovia*) has found the presence of a small amount of essential oils which is similar to the essential oils found in *Smirnova iranica*.

Table 3. Chemical ingredients comprising the essential oil of the leaf of the *Smirnova iranica* sabeti

Number	Name of compound	RT	Percent compound of site			
			1	2	3	4
1	4-Heptanone, 3-methyl-	13.87	-	-	3.81	-
2	2-Cyclopenten-1-one, 2-hydroxy-	18.32	3.44	0.68	-	-
3	5,9-Undecadien-2-one, 6,10-dimethyl-, (E)-	28.27	3.41	1.30	5.87	2.20
4	trans- β -Ionone	29.78	7.76	9.24	19.06	10.59
5	4-Hexen-3-one, 5-methyl-	50.97	-	-	3.54	3.46
6	(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-	31.85	5.84	2.03	4.42	2.57
7	2-Pentadecanone, 6,10,14-trimethyl-	46.93	2.16	-	4.47	3.47
8	11,14,17-Eicosatrienoic acid, methyl ester	51.95	-	0.48	4.80	-

Other studies, have not found any essence in plants of the same subcategory of this plant.

As is shown in Table 3, the leaves of *Smirnova iranica* contain 8 different compounds with different amounts at all sites. Number of compounds in sites 1 and 3 was 12 and for the sites 2 and 4, it was 11 and only in site 3, all detected compounds were above 3 percent.

Among these compounds, 5,9-undecadien-2-one, 6,10-dimethyl-, (E)-trans- β -Ionone, dodecanoic acid, (4H)-benzofuranone,

5,6,7,7 a-tetrahydro-4 , 4,7 a-trimethyl-, tetradecanoic acid, hexadecanoic acid and phytol were the seven common compounds found in the leaves of all sites. The ingredient of 4-heptanone, 3-methyl- with 3.81 % only occurred in 3 sites.

Hexadecanoic acid ingredient (10.20 %), trans- β -ionone (7.76 %) and Vitamin E acetate (7.21 %) were the three main and ingredients of the site 1. At site 2 tetradecanoic acid (20.57 %), hexadecanoic acid (14.82 %) and (Z, Z, Z) -9,12,15-

octadecatrien-1-ol, (12.47 %) were ranked first to third, respectively. In sites 3 and 4, two ingredients of trans- β -ionone with 19.06 % and 10.59 % and dodecanoic acid with 11.93 % and 7.49 % were respectively identified as the main ingredients of these sites. Hence we can say that the main compounds identified in the leaf samples were compounds such as trans- β -ionone, dodecanoic acid, tetradecanoic acid, hexadecanoic acid and phytol which were more than 3 percent.

CONCLUSION

Trans- β -ionone, dodecanoic acid, tetradecanoic acid, hexadecanoic acid and phytol have the most amount in leaf that is reported by M.Ghavam in 2015. Tetradecanoic acid (acid myristic), hexadecanoic acid (palmitic acid) and dodecanoic acid (lauric acid) are saturated fatty acids. Myristic acid is used as sunscreen, softener, moisturizer and cleanser. Palmitic acid is a fatty acid, solid and waxy with a melting point of 64 ° C which is found as palmitin in solid fats, palm oil, and natural fats, including milk fat. This acid has lubricant, binder and defoamer properties. Lauric acid appears as a white crystalline that eliminates the bacteria; the bacterial infections caused by intestinal

parasites and infections of a variety of diseases ranging from AIDS to common colds. In fact, the human body converts lauric acid into a material called monolaurin which is an antibacterial and antiviral agent, especially for viruses with a lipid envelope such as HIV and influenza. Aliphatic alcohols lower the cholesterol levels and help to prevent platelet aggregation. Phytol which is an alcohol of this type, has antioxidant and anticancer benefits. β -Ionone reduces the activity of HMG-CoA reductase of the liver.

Hence, it can be concluded that *Smirnova iranica* is a versatile plant found in sandy fields of Iran with medicinal, anti-virus, anti-cancer and anti-bacterial properties and is rich in vitamin A precursor. Also, in terms of selecting the sites, site 2 was clearly more privileged than the others, both quantitatively and qualitatively. Therefore, site 2 can be regarded as the optimal point of harvest.

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