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A Study On The Change In The Components Of Volatile Oil Of *Salvia* Farinacea Grown In Different Environments

Hasan Basri KARAYEL[1]
[1]Dumlupınar University. Gediz Vocational School, Medical and Aromatic Plants Department. Kütahya, Türkiye.
kbasri23@hotmail.com

***Abstract*—This study was conducted simultaneously in three different locations, which are Çanakkale, Balıkesir and Kütahya, in order to define the effect of location on the volatile oil components, volatile oil rate and volatile oil quality in a Medical Sage species (Salvia farinacea)in the years 2015-2016. Field experiments were repeated in 3 replicates according to randomized block design. These plants' volatile oils were obtained by hydrodistillation method (GC_MS/FID) and the avarage of two years' volatile oil rates in three different locations were measured as %0.01, %0.01, %0.0 respectively. The basic components of the volatile oil were determined as follows in Çanakkale in 2015 and 2016 respectiely; β-caryophyllene %4,50 and %18,92, Germacrene-D %10,51 and %10,82, β-bisabolene %10,24 and %11,74. In Balikesir, the numbers of main components are as follows again for the years 2015 and 2016 respectively; β-caryophyllene %10,64 and %15,97, Germacrene -D %10,49, and %7,78, β-bisabolene %0 and %13,63. No volatile oil was obtained in the first year and the plants were damaged by cold weather conditions in the second year in Kütahya. As a result of the study it was found out that volatile oil components are rich in terpenes and the amount of volatile oil differs according to the ecological factors.**

Keywords—Salvia farinacea, Environment, Essential oil

1- Introductıon

Salvia, which has been known since ancient times and has never lost its importance to this date, constitutes an important part of the medicinal plants. The first records of its medicinal properties and the fact that this plant was used in the treatment of various diseases have been found in the ornamented writings and paintings of the ancient tombs and monuments. Due to its healing properties, it is called *Salvia*, which comes from the word Salveo which means "Save and heal". Monoterpenes and their oxygenated derivatives in the composition of the essential oils of the species have antiseptic effect. [8]. Terpenes form the largest group of chemical structures of essential oil compositions. However, there are also small amounts of alcohols, aldehydes, esters, phenols, nitrogen and sulfur-containing compounds. Oxygen derivatives resulting from oxidation of terpenes are substances that have odor, taste and therapeutic properties [5]. Essential oils have been widely used in Roman, Greek and especially Egyptian civilizations. In recent years, the interest in aromatherapy, a branch of alternative medicine, has also increased the use of essential oils. Etheric oils are used in massage treatments or in relaxing baths [4].

Salvia species are used not only in the medical field, but also as a sweetener in the perfumery and food industry [2]. Most Salvia species are cultivated as decorative ornamental plants in parks and gardens due to their beautiful appearance [1-9]. Although there are numerous studies on the chemical properties of the genus, the morphological and anatomical studies that help the introduction of the genus are limited [11]. The reasons such as the fact that the synthetic origin substances produced in recent years have more side effects and especially the resistance of organisms against synthetic drugs used as antimicrobials, have increased the importance of natural plant sources and the medicinal plants carrying these substances [10]. Considering that the volatile oil components differ according to environmental factors, In order to discover the chemical structure of *Salvia species*, it is needed to carry out more studies on different *Salvia species* from different locations of our country, which is rich in *Salvia species* [17]. Our aim in this study is to bring different types of sage into the economy.

2- Material and Method

2.2 Establishment of Plant Material and Experiments

The seeds used as material in this study were obtained from Ankara University, Faculty of Agriculture, Department of Field Crops. In December 2014, seeds were planted in the viols of Hekim Sinan Medicinal Plants Botanical Garden of Kutahya Municipality to make seedlings. The research was conducted simultaneously in the years 2015-2016 in three different locations which are "Çanakkale Onsekiz Mart University, Faculty of Agriculture, Dardanos Campus trial area", "Balıkesir Edremit Kale Natural Corporation, Medical Plants Garden" and "Kütahya Municipality Hekim Sinan Medical Plants Botanical Garden. As 180 plants were needed in each location, 216 seedlings were planted taking the

possible failures in the greenhouse and the field process into account. The viols have been given water with a strainer bucket at regular intervals. The germination of the seeds lasted 20-25 days. The rooted seedlings began to be moved to the field in April 2015. Seedlings were given water of life after they were moved to the field. Field trials were carried out in 3 replications, according to random block trial design. Plant planting frequency is arranged according to 50 cm horizontal, 30 cm vertical distances and each parcel consists of 3 rows. They were harvested once in the fİrst year and three times in the second year in all locations and the flowering season was preferred as the harvest time which is the best time to get maximum amount of essential oil.

2.3 Essential Oil Production

The amount of essential oil in the determination of the amount of essential oil of spice medicines, condiments and medicinal plants is all substances which are vaporized by the conditions specified in the method and expressed in milliliters per 100 g of dry matter. T he reagents used must be in analytical purity and the pure water used must be distilled or equivalent purity. This method is based on the principle of distilling the aqueous suspension of the sample, collecting the discrete volatile oil in the distillate fraction from the aqueous phase, collecting the volatile oil collected, and then calculating the percent volatile oil. At the beginning of the test volatile oil analysis 20 g dry material was weighed. The material was taken into a 500 ml balloon with a round base and a shaved neck. 200 ml (may vary depending on the amount of sample, about 10 times), added with pure water and shaken. The balloon is placed on the heated jacket of the Neo-Cleaver system, the lower part of the vertical glass tube is connected to the glass balloon, and the upper part is connected to the cooling system. After filling the tube and the slant pipe with pure water, the coolant system is started and the balloon is heated. It is hydrodesticated for 2 hours. After the system has cooled down and the volatile oil collected in the graduated fraction has separated from the aqueous phase, the amount (ml) is determined. The amount of essential volatile oil in 100 g is calculated as the volatile oil content (%) according to the amount of sample (g) to be weighed [15].

2.4- Determination of essential oil composition by GC-MS

Analyzes of essential oil components were carried out at the Research Laboratory of the Western Mediterranean Agricultural Research Institute. Samples were diluted with 1% hexane and injected in 1 µl with 40:1 split ratios to Gas chromatography (Agilent7890A). Capillary columns (HP InnowaxCapillary; 60.0 m x 0.25 mm x 0.25 µm) were used to separate the components. The column was split into two fractions at a rate of 1:1 using a splitter to the FID and mass spectrometry detector (Agilent5975C). In the analysis, helium was used as carrier gas at a flow rate of 0.8 ml / min. The injector temperature was maintained at 250 ° C, the column temperature program was 10 minutes at 60 ° C, 4 ° C / minute (40 minutes) at 60 ° C and 220 ° C and 10 minutes at 220 ° C It is set to be 60 minutes. The scan range (m / z) for the mass detector is 35-450 atomic mass units and the electron bombardment ionization energy is 70 eV. The diagnosis of volatile oil components is based on the data from Oıl Adams, Wıley and Nıst libraries. The data of the FID detector were used for the volatile oil component ratios[12].

3. Findings and Discussion

The average yields of *Salvia farinacea* species which was examined in this study were measured 0.01% in Çanakkale, 0.01% in Balıkesir (Edremit) and 0.0% in Kütahya.

Table 3.1. the amount of components of the essential oil of Salvia *farinacea* species according to locations and years (%)

		Çanakkale		Balıkesir		Kütahya	
Sequence	Component	1st year	2nd year	1st year	2nd year	1st year	2nd year
1	Limonene	-	-	13,15	-	-	Cold Damage
2	1-octadecanol	-	5,33	3,43	18,66	-	-
3	viridiflorol	-	7,54	3,02	5,79	-	-
4	Cymene	-	-	6,63	-	-	-
5	Tridecane	-	-	1,72	-	-	-
6	Tetradecane	1,99	-	7,05	-	-	-
7	α-thujone	2,44	-	-	-	-	-
8	Octen – 3- ol	2,04	-	-	-	-	-
9	β-thujone	2,37	-	-	-	-	-
10	Pentadecane	3,13	-	11,62	-	-	-
11	Camphor	2,94	5,33	2,76	2,42	-	-
12	α-terpinolene	-	-	2,06	-	-	-
13	Hexadecane	2,14	-	8,72	-	-	-
14	β-caryophyllene	4,50	18,92	10,64	15,97	-	-
15	Heneicosane	-	-	1,8	-	-	-
16	α –humulene	1,91	8,4	2,62	4,47	-	-
17	Heptadecane	-	-	2,94	-	-	-

18	α-terpineol	1,99	-	-	-	-	-
19	Germacrene -D	10,51	10,82	10,49	7,78	-	-
20	β - bisabolene	10,24	11,74	-	13,63	-	-
21	δ - cadinene	3,55	5,85	3,40	2,33	-	-
22	Caryophylleneoxide	5,18	14,29	2,91	3,52	-	-
23	2-pentadecanone	4,33	5,17	2,4	2,39	-	-
24	Carvacrol	3,48	-	2,25	7,73	-	-
25	Manool	20,59	-	-	-	-	-
26	Hexadecanol	9,63	-	-	-	-	-
27	Phytol	5,04	-	6,25	-	-	-
	Total	100	99,64	99,61	98,93		

While the average values of the species were determined by the samples taken from only one harvest in the first year, they were determined by the samples obtained from 3 different harvests. There was no significant difference between the samples obtained from three harvests in the second year. The avarages of all the harvests of *Salvia farinacea* are shown in the table 3.1. for different locations. The main components of the *Salvia farinacea* species are β-Caryophyllene, Germacrene D, β-bisabolene and they were determined as the main components in all three locations. The proportions of the main components by location are given below respectively. The main components of the volatile oil of *Salvia farinacea* plant were determined as follows for the year 2015 according to different locations; β-Caryophyllene 4,50%, Germacrene-D 10,51%, β-bisabolene 10,24% in Çanakkale; β-Caryophyllene 10,64%, Germacrene-D 10,49%, β-bisabolene 0% in Balıkesir and no volatile oil was obtained in Kütahya in 2015. The main components of the volatile oil of *Salvia farinacea* plant were determined as follows for the year 2016 according to different locations; β-Caryophyllene 18,92%, Germacrene-D 10,82%, β-bisabolene 11,74% in Çanakkale; β-Caryophyllene 15,97%, Germacrene-D 7,78%, β-bisabolene 13,63% in Balıkesir and the plants were damaged by cold weather conditions in Kütahya in 2016. The total percentages of component numbers and the components that were determined in *Salvia farinacea* species are listed as follows: In Çanakkale 19 components and 100% essential oil in the first year, 11 components and 99.64% essential oil in the second year; in Balıkesir 19 components and 99.61% essential oil in the first year, 12 components and 98,93% essential oil in the second year. Other important components of this species have also varied. The important components were determined as Phytol, Manool, Caryophylleneoxid for Çanakkale location and Pentadecane, 1-octadecanol, Phytol for Balikesir location. The essential oil components of the species varies to some extent according to the location. β-Caryophyllene was determined as the main component of *Salvia farinacea* species with the highest rate in both locations. This shows how effective the ecological factors are on the essential oil components. According to the research results, factors such as growing environment and number of harvests can be said to be effective on volatile oil components. In addition, the variety and quantity of bioactive substances present in medicinal and aromatic plants may also vary according to the part of the plant used, the post-harvest processes, and the methods for obtaining volatile oils and methods of analysis [14]. Similar to our study, seven *Salvia species* were collected in the Dallas Arboretum botanical garden. These species (*Salvia coccinea*, *S. farinacea*, *S. greggii, S. leucantha, S. longispicata* × *farinacea, S. medrensis, S. roemeriana and S. splendens species*) were investigated by microdistillation technique. The volatiles were obtained by GC / MS method. Seven compounds were identified representing 94.3 - 99.7% of the fats. The main components common in all the seven species were found as; *S. farinacea* 1-octene-3-ol (30%) and (Z) -3-hexene (23%); longispicata × farinacea 1-octene-3-ol (50%) and (Z) -3-hexenal (24%); S. medrensis (Z) -3-hexenal (53%); S. Roemeriana limonene (49%) and α-pinene (20%) [16]. In another study of *S. farinacea*, the blue sage, which is called Raider Azure and which is a natural ornamental plant which is resistant to drought, was investigated. In the essential oil obtained from *S. farinacea*, terpene, camphor, β-pinene, α-pinene, cineole, camphene components were obtained as the main constituents [7]. In a study conducted in Romania, eight species of *Salvia L.* (*S. amplexicaulis, S. austriaca, S. farinacea, S. pratensis, S. sclarea, S. splendens, S. transsilvanica and S. Verticillata*) were analyzed by GC / MS. . The resulting components (β-pinene and D-germacren) were the most common chemical constituents of *Salvia* oils in different percentages [13]. Natural wildflowers are a rich source of genes for continuous plant growth. Texas Tech University has introduced a species of bluish “Raider Azure” and blue sage (*Salvia farinacea* and *Salvia farinacea* Benth.). The university has made its cultivation as a natural ornamental plant resistant to drought. The main components of the volatile compounds obtained in this study were camphor, α-pinene, β-pinene, cineole, camphene [6]. In the study conducted in Egypt, the aim was to determine the volatile oil content and the essential oil content of *Salvia farinacea* Benth. The main components of the leaf were allo-aromadendren (19.35%) and aromadendren (18.3%) and then selin-3,7 (11) -dien (10.2%), 18-cadinene (18.56%) and aromadendren (17.94%), and then β -myrcene (10.5%) and E-β-

okimen (9.11%). The main constituents of flowers were 1-octen-3-ol (46.51%), 2-hexenal (18.02%), benzaldehyde (8.8%) and aromadendrene (7.24%) [3]. The data obtained by many researchers and *Salvia farinacea* values are not similar. The main reason for this may be ecological factors.

4- Results

In the studies carried out in Çanakkale, Balıkesir, Kütahya locations, the volatile oil components of Salvia farinacea, which were grown in regions with different soil structure and climatic characteristics, varied. The main constituents of Salvia farinacea were found to be β ,6 caryophyllene 4,50-18,92% Çanakkale, 10,64-15,97% Balıkesir in the first and second years, 1-octadecanol in the second year in Balıkesir, 18,66%, In the first year, Manool was identified as 20.59% in Manisa and 10.24-11.74 in β-bisabolene in Çanakkale in the first and second years. The most volatile oil component was determined in Balıkesir with 19-12 components. In this study, the amount of essential oil and the changes in the main component in the volatile oil varied depending on the genetic and environmental factors.

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