

Examination Of The Change In The Components Of Volatile Oil Of *Salvia Virgata* Jacq. Plant Grown In Different Locations

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Abstract—This study was conducted in three different locations (Çanakkale, Balıkesir and Kütahya) in order to define the effect of location on the volatile oil components, volatile oil rate and volatile oil quality of *Salvia virgata* Jacq. plant simultaneously in the years 2015-2016. Field experiments were repeated in 3 replicates according to randomized block design. The seedlings were planted by a horizontal distance of 30 cm and vertical distance of 50 cm. These plants' volatile oils were obtained by hydrodistillation method (GC_MS/FID) and the average volatile oil rates of the two years in three different locations were measured as %0.01, %0.02, %0.02, respectively. While no major component was found in 2015, the main components of the volatile oil of *Salvia virgata* Jacq. plant were determined as follows for the year 2016 according to different locations: for Çanakkale location β -Caryophyllene %29,42, Germacrene-D %9,92, Caryophylleneoxide %26,92; for Balıkesir location β -Caryophyllene %48,12, Germacrene-D %9,75, Caryophylleneoxide %13,22 and for Kutahya location β -Caryophyllene %39,43, Germacrene-D %14,43, Caryophylleneoxide %6,94. While no components were obtained from *Salvia virgata* Jacq. plant in the first year in all three cities, 10 components and 84,23% volatile oil components were obtained in Çanakkale, 21 components and 99,9% volatile oil components were obtained in Balıkesir and 18 components and 98,32% volatile oil components were obtained in Kütahya in the second year. Some other important components of this species also differ according to location. As a result of the study it was determined that volatile oil components are rich in terpenes and the amount of volatile oil differs according to the ecological factors.

Keywords—*Salvia virgata* Jacq., Volatile oil, Volatile oil components

1- Introduction

The *Salvia* genus, which belongs to the Lamiaceae (Labiatae) family, contains over 98 species of both species in the world, spread to the Mediterranean and Central Europe, especially in the tropics and subtropics. 106 of these species are naturally

distributed in our country and 58 of them are endemic. Turkey is ranked 13th in the world in terms of species richness of *Salvia* species [3].

It is a very branched and perennial herbaceous species that can reach 80-100 cm in height. Its flowers are in dark purple color, the leaves are at the bottom and in the form of a rosette, it has handle and a long egg shape. It is a common plant in Turkey. It was found that the flower parts contained 3% volatile oil. The leaves are used externally as ointment for wound [1].

Salvia virgata Jacq, a species belonging to the genus *Salvia*, is a perennial plant that can grow up to 20-160 cm. and it prefers a wide variety of habitats such as bushes, forests, meadows, empty fields, limestone and volcanic rocks and roadside. It is distributed from sea level up to 2300 m. Its flowering time is between May and September. *S. virgata* Jacq., which is distributed in the Crimea, the Balkans, Italy, the Caucasus, Northern Iraq, Iran, Afghanistan and Central Asia, is also distributed in Turkey and on islands [6].

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]. In recent years, the interest in aromatherapy, a branch of alternative medicine, has also increased the use of essential oils. Etheric fats are used in massage treatments or in relaxing baths [7]. This study was carried out in order to determine the amount of essential oil and the change in the essential components of the essential oil of sage in different locations. This research is a study on the determination of essential oils of the *S. virgata* Jacq plant belonging to the genus *Salvia* L. in Aegean and Marmara region. It was also carried out for the comparison of essential oil compositions of *Salvia* L. species and the evaluation of its essential oils.

2- Material and Method

2.1 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 fields 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 first 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.

2.2 Essential Oil Production

At the beginning of the test volatile oil analysis 20 g dry material was weighed. The material was taken into a 500 ml balloon. 200 ml of pure water (may vary depending on the amount of sample, about 10 times of the material) was added and shaken. It was hydrodesticated for 2 hours and volatile oil was acquired. After the system has cooled down and the volatile oil was extracted from the aqueous phase, its amount (ml) was measured. The amount of essential volatile oil in 100 g was accepted as the volatile oil rate (%) according to the amount of sample (g) weighed. [13].

2.3 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 (HPInnowaxCapillary; 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

(Agilent 5975C). 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 was set to be 60 minutes. The scan range (m / z) for the mass detector was 35-450 atomic mass units and the electron bombardment ionization energy was 70 eV. The diagnosis of volatile oil components is based on the data from Oil Adams, Wiley and Nist libraries. The data of the FID detector was used for the volatile oil component ratios [11].

3. Findings and Discussion

The average yields of *Salvia virgata* Jacq. species which was examined in this study were measured 0.01% in Çanakkale, 0.02% in Balıkesir (Edremit) and 0.02% in Kütahya. 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 averages of all the harvests of *Salvia virgata* Jacq. are shown in the table 3.1. for different locations. The main components of the *Salvia virgata* Jacq. species are β-Caryophyllene, Germacrene D, Caryophylleneoxide and they were determined as the main components in all three locations. The proportions of the main components by location are given below respectively. While no major component was found in 2015, the main components of the volatile oil of *Salvia virgata* Jacq. plant were determined as follows for the year 2016 according to different locations: for Çanakkale location β-Caryophyllene %29,42, Germacrene-D %9,92, Caryophylleneoxide %26,92; for Balıkesir location β-Caryophyllene %48,12, Germacrene-D %9,75, Caryophylleneoxide %13,22 and for Kutahya location β-Caryophyllene%39,43, Germacrene-D %14,43, Caryophylleneoxide%6,94. While no components were obtained from *Salvia virgata* Jacq. plant in the first year in all three cities, 10 components and 84,23% volatile oil components were obtained in Çanakkale, 21 components and 99,9% volatile oil components were obtained in Balıkesir and 18 components and 98,32% volatile oil components were obtained in Kütahya in the second year.

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

Sequence	Component	Çanakkale		Balıkesir		Kütahya	
		1 st year	2 nd year	1 st year	2 nd year	1 st year	2 nd year
1-	α-thujone	-	-	-	0,82	-	-
2-	Sabinene	-	-	-	2,02	-	1,07
3-	γ-Terpinene	-	-	-	1,03	-	-
4-	α-Thujone	-	-	-	0,58	-	-

5-	α -Cubebene	-	-	-	0,64	-	0,55
6-	α -Copaene	-	-	-	2,03	-	1,94
7-	Linalool	-	-	-	0,73	-	-
8-	Linalylacetate	-	-	-	0,64	-	-
9-	β -Copaene	-	-	-	0,72	-	0,74
10-	β -caryophyllene	-	29,42	-	48,12	-	39,43
11-	α -Humulene	-	2,66	-	2,83	-	2,52
12-	γ -Muurolene	-	4,35	-	3,83	-	3,89
13-	Germacrene D	-	9,92	-	9,75	-	14,43
14-	β -Bisabolene	-	1,53	-	1,14	-	2,82
15-	δ -cadinene	-	2,21	-	3,6	-	3,83
16-	γ -Cadinene	-	-	-	1,54	-	1,63
17-	Caryophylleneoxide	-	26,92	-	13,22	-	6,94
18-	Humuleneepoxide-II	-	1,75	-	0,74	-	-
19-	Spathulenol	-	-	-	0,84	-	2,83
20-	14-Hydroxy β caryophyllene	-	4,14	-	1,35	-	0,94
21-	viridiflorol	-	1,83	-	-	-	-
22-	carvacrol	-	-	-	-	-	4,41
23-	phytol	-	-	-	-	-	2,56
24-	β -ylangene	-	-	-	-	-	0,54
	Total		84,23		99,9		98,32

The important components were determined as 14-Hydroxy- β -caryophyllene, γ -Muurolene, α -Humulene for Çanakkale location; δ -Cadinene, γ -Muurolene, α -Humulene for Balıkesir location and carvacrol, γ -Muurolene, δ -Cadinene for Kütahya 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 virgata* Jacq. species with the highest rate in all three 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. Similar to our study, it was determined that there are 29 compounds in the volatile oil of *Salvia virgata* which constitutes 98.36% -99.18% of the volatile oil of these compounds in a study. Essential components of essential oil were determined as β -Caryophyllene (24.58% -42.54%), Caryophylleneoxide (10.25% -19.88%), Sabinene (8.64% -19.58%), 1-Octen-3 -Ol (7.54% -8.59%), terpinene-4-ol (4.25% -6.64%) and α -thujone (3.74% -6.46%) [2]. It was found out that *Salvia virgata* has 23 compounds in its volatile oil and these compounds are β -Caryophyllene (35.2%), (Z) - β -farnesen (10.1%), Caryophylleneoxide (6.1%) and α -pinene (%) 5,7) [4]. The chemical composition of four *Salvia* species from Turkey (Bentham *S. trichocla.*, *S. Virgata* Jacq., *S. Ceratophyllum* L., *S. multicaulis* Vahl.) were identified by GC-MS (gas chromatography- mass spectrometry) system. The investigators identified 40, 40, 3 and 39 compounds representing 91.9%, 90.4%, 89.7% and 88.4% of *S. trichoclada*, *S. virgata*, *S. ceratophylla* and *S. multicaulis* oils respectively. Caryophylleneoxide (25.1%), spathulenol (15.4%) and β -pinene (12.3%) were the main components of *S. trichoclada*. They determined the main compounds of *S. virgata* as 1,8-

Cineol (20,3%), α -copaen (18,6%) and germakren-D (17,6%). The main components of *S. multicaulis* were Caryophylleneoxide (22.5%), spathulenol (12.7%) and β -pinene (7.5%) [8]. The composition of the volatile oil obtained from the dried flowered portions of *Salvia virgata* was analyzed by GC and GC-MS. As a result of the analysis 8 components were found in the essential oil of the *Salvia virgata* Jacq. species. The essential components of essential oil are Caryophyllene Oxide (34.4%), spathulenol (25.6%), 1-docosanol (11.7%), n-tetradecane (9.3%) and geranyl acetone (5.6%) [9]. The essential oils in the plant parts were analyzed in a study conducted on *Salvia virgata* Jacq. ve *Salvia syriaca* L. species collected from wild habitat. As a result of the analysis, the essential components of the species *Salvia virgata* Jacq. were found to be β -caryophyllene (46.6%), Germacrene (13.9%), Caryophylleneoxide (13.2%), spathulenol (6.4%) and Germacrene. D (5.7%) [12]. According to the results of many researchers, the volatile oil components of the species *Salvia virgata* Jacq. are similar.

4- Results

In the studies carried out in Çanakkale, Balıkesir and Kütahya, *Salvia virgata* Jacq. species differed in terms of essential oil components. β -Caryophyllene, Germacrene-D, Caryophylleneoxide were determined as the main components of *Salvia virgata* Jacq. and similarly, β -Caryophyllene, Germacrene-D, Caryophylleneoxide were also determined as the main component in three locations. β -Caryophyllene was determined as the main component with the highest rate in all three locations. In order to discover the chemical structure of *Salvia* species, it would be useful to carry out more studies on different *Salvia* species from different locations of our country, which is rich in *Salvia* species.

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