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## Volatile oil studies of some aromatic plants in Taif region

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This study was carried to investigate the volatile oil content of (basil - rosemary - marjoram - rose) plants grown in different regions at Taif Governorate. Moreover, the main components of volatile oil as well as, changes in its compositions were also investigated as a result of changing the collection regions. The samples for volatile oil extraction were collected from several regions and volatile oil was extracted and consequently its percentages were calculated. The volatile oil composition was occurred by using GC-MS and the main components of volatile oil from different regions were compared. The results of this study showed that the volatile oil percentage was changed among the collection samples in all studied plants. Moreover, the percentages of the main components of volatile oil were also differed as results of changing the geographical regions. Studying the environmental conditions of the location which aromatic plants will be cultivated is recommended according to our study.

Keyword: Volatile Oils; Rose; Rosemary; Marjoram; Basil; Oil Composition.

#### **1. Introduction**

Herbs play an important role in maintaining human health and volatile oils have been of great interest due to their antioxidant capacities. Volatile oils also have been the sources of natural products and used for many medical products. Volatile oils are plant secondary metabolites that are known for their fragrance and food flavour properties. They consist of a complex mixture of mono and sesqui-terpenes, phenyl propanoids and oxygenated compounds<sup>[56]</sup>. The main advantage of natural agents is that they do not enhance the resistance", "antibiotic a phenomenon encountered with the long term use of synthetic antibiotics<sup>[62]</sup>. Essential oils offer a promising potential for future applications within the fields of agriculture, medicine, pharmaceutical industry and biotechnology because of its function as signaling compounds between different types of organisms and diverse biological systems, their general antimicrobial and antioxidative effects and medicinal activity.

Despite the advances in new pharmaceuticals, many products sold by drug companies are still plants. Most herbal medicines of current interest originate from the ancient civilizations of Africa, the Asian subcontinent, and North, Central and South America<sup>[51]</sup>. Production, composition and quality characteristics of essential oils from basil, marjoram, rosemary and rose have been investigated<sup>[25, 28, 31, 41, 55, 59, 60, 72, 74]</sup> and cultivation guidelines stressing geographic and climatic needs have been devised<sup>[20]</sup>.

Lamiaceae family is known for the wealth of species with medicinal properties, which have been used since early times and many of these species are common in Mediterranean region<sup>[2]</sup>. The Lamiaceae plants are generally aromatic in

all parts including a number of widely used culinary herbs, such as rosemary, basil and marjoram<sup>[16, 29,73]</sup>. Sweet basil (Ocimum basilicum L.), the oldest spices belonging to the Lamiaceae commercially and family. is extensively cultivated for essential oil production in many continents around the world [8]. Basil leaves containing essential oils of distinctive aroma can be used both fresh and dried to spice up various kinds of meals. Apart of culinary use, basil has been traditionally employed as a medicinal herb in the treatment of headaches, coughs, diarrhoea, constipation, warts, and/or kidney malfunction<sup>[27,</sup> <sup>48, 53]</sup> and basil has been extensively utilized in food and perfumery industries <sup>[64]</sup>. Recently the potential uses of basil essential oil, particularly as antioxidant and antimicrobial agents have also been explored <sup>[14, 29,43, 53]</sup>. Even the smell of basil is considered to be curative <sup>[6]</sup>. Linalool, methyl chavicol, 1,8-cineole, eugenol, and methyl cinnamate were the major components in different varieties of sweet basil volatile oil [30, 36, 72].

Rosmarinus officinalis L. (Rosemary) is a very important medicinal and aromatic plant, which widely used in folk medicine, culinary, cosmetic virtues and for the flavouring of food products <sup>[52]</sup>. Rosemary essential oil is of immense medicinal worth for its powerful antimutagenic, antiphlogistic, antioxidant, chemo-preventive and properties, anti-inflammatory, antibacterial antiseptic, antispasmodic and anti-diabetic [1,16, 35, <sup>60]</sup>. The volatile oil of rosemary reaches to 1.43 % <sup>[74]</sup> with the main component of 1,8-cineole. In the previous work dealing with chemical composition of rosemary essential oil,  $\alpha$ -pinene is reported as the major component, followed by 1,8-cineole, camphene, ß-myrcene, camphor and borneole <sup>[31, 45, 25]</sup>. The results of GC-MS analysis of rosemary volatile oil show that the main components of volatile oil were  $\alpha$ -pinine, camphine, 1,8-cineol, verbinone and borneol <sup>[29]</sup>. Interestingly, they also added that the camphor component was not detected in the oil composition although different authors observed it <sup>[60, 74]</sup>.

Origanum majorana, L. (Marjoram) plants are extensively used for the flavoring of alcoholic

beverages, food products and in perfumery due to their spicy fragrance <sup>[24,47]</sup>. This genus includes some important culinary herbs, commercially available and exportable plants with appreciable market values <sup>[11, 22]</sup>. It has been more interest regarding the biological properties of marjoram essential oil because of its antimicrobial and antioxidant potential <sup>[3, 19, 49, 57]</sup> and possesses high antioxidant and anticancer properties <sup>[55]</sup>. Essential oil content of marjoram ranged from 0.57% to 2.00 % <sup>[41]</sup>. The main constituents of commercial marjoram were terpinen-4-ol, cissabinene hydrate and  $\gamma$ -terpinene. The main identified oils constituents of Majorana hortensis were  $\gamma$ -terpinene, sabinene hydrate, terpinen-4-ol,  $\alpha$ -terpinene and sabinene <sup>[25]</sup>.

Damask rose (Rosa damascena Mill.) is the most important source of rose products such as rose oil, rose concrete, rose absolute and rose water <sup>[7,</sup> <sup>63]</sup> and rose oil obtained from it is traditionally preferred <sup>[59]</sup>. Roses are highly expensive base materials for natural perfume, the fragrance, flavor, cosmetic, pharmaceutical and food industries. Rose oil is a very expensive essential oil, partly due to the lack of quality natural and synthetic substitutes <sup>[63]</sup>. Rosa damascena Miller is one of the most important commercial crops in Taif region in Saudi Arabia and the gulf region <sup>[9]</sup>. Although Saudi Arabia is not the most hospitable of lands for flowers, they flourish in Taif<sup>[12]</sup>. Taif rose oil and rose water have become important commercially valuable products [38].

Essential oil from the rose is reported to have analgesic, antispasmodic effects, anti bacterial and hypnotic activities <sup>[10, 40, 54]</sup>. Taif rose oil is supposed to help in the treatment of depression, to help reduce tension and stress. It is meant to cure headaches and insomnia, is considered a powerful sexual tonic and of great help during the menopause. The essential oil content of rose varied from 0.032% to 0.049% and that of hybrid roses from 0.037% to 0.05% <sup>[37]</sup>. Citronellol, geraniol, linalool, nerol and 5-methyl octadecane were reported to be the main constituents of Taif rose oil <sup>[9, 37, 38,58, 69]</sup>.

There are many reports in the literature showing the variation in the yield and chemical composition of the essential oil with respect to geographical regions and altitude [4, 16, 50, 61, 68]. Climatic factors such as heat and drought were also related to the essential oil profiles obtained <sup>[44, 67]</sup>. The chemical composition of basil volatile oil was varied according to the plant growing location and some components were disappeared in some locations and presented in the others [76]. <sup>[34]</sup> suggested that the climate of the zone plays an important role at the essential oil level production.<sup>[75]</sup> reported that rosemary essential oil yield was increased in bioclimatic zones with lower thermicity indexes and the essential oil chemical composition undergoes variations over the different bioclimatic areas studied. [74] found that the yield and chemical composition of essential oil of marjoram from twenty three localities were significantly varied. Depending on the locations where the plants grew, the essential oil yield and composition were significantly differed <sup>[67]</sup>.

There is limited published research on basil, rosemary, marjoram and rose growing criteria despite their popularity and their several uses. In addition, there is a lack in the information concerning the comparison of volatile oil composition among different growing locations in Taif region despite of the suitable conditions for volatile oil production. Therefore, the aim of this project was to study the changes in volatile oil content of basil, rosemary, marjoram and rose grown in different locations in Taif Governorate.

## 2. Materials and Methods

This study was carried out at Faculty of Science, Taif University, Taif, Saudi Arabia during 1430 and 1431 seasons. The plants used in this study were basil-rosemary -marjoram-rose. The plant samples were collected from four regions in Taif governorate i.e. Tarbah, Ranih, Al-Hada and Al-Haweiah regions while rose samples were collected from Al-Shafa, Al-Hada and Al-Haweiah regions. From each region, five samples of each plant were taken and transported to the laboratory of Faculty of Science, Taif as soon as possible for volatile oil extraction.

## 2.1. Volatile oil extraction method

The volatile oil percentages in all samples obtained from each replicate of every location were determined by a water distillation method described in <sup>[15]</sup> using the following equation:

Volatile oil percentage = oil volume in the graduated tube / fresh weight of sample x 100.

## **2.2. Volatile oil composition**

The obtained volatile oil was dehydrated over anhydrous sodium sulphate and stored in refrigerator until GC-MS analysis. Essential oil samples were performed using a Varian GC CP-3800 and MS Saturn 2200 equipped with a Factor Four capillary column (VF-5ms 30 X 0.25 mm ID and film thickness 0.25 µm). An electron ionization system with ionization energy of 70 eV was used for GC-MS detection. The carrier gas was helium at a flow rate of 1 ml/minute. The tempmerature program for rosemary, marjoram and basil was as follows: The MS transfer line temperatures of injector and detector were set at 140 and 300 °C, respectively. The initially column temperature was 50 °C at 2 °C/minute and increased to 260 °Cat 5 °C/minute, finally it hold at 260 °C for six minutes. A sample of 1 µl (spilt ratio 1:40) from volatile oil was manually injected.

Concerning volatile oil analysis of rose, the temperature program was as follows:

The oven temperature was programmed for 5 minute at 60  $^{\circ}$ C, 60  $^{\circ}$ C to 290  $^{\circ}$ C at 6 $^{\circ}$ C/ minute and held for 5 minute finally at 290  $^{\circ}$ C, solvent delay time 3 min. The injection of the all the samples were carried out with the auto-sampler for 1 µl with a split ratio 1/40. The conditions of analysis and specification of the instrument were optimized for better separation and resolution. The volatile oil components were identified by comparing their retention times and mass spectrum with those of standards, NIST library of the GC-MS system and literature data.

## 3. Results

#### 3.1. Basil volatile oil

The chemical composition of basil volatile oil obtained from four locations in Taif governorate was presented in Table (1). The volatile oil percentage was varied from 0.68 to 0.79 %

according to the sample location. The GC-MS results showed that the main components of volatile oil were 1,8-cineole, Linalool, Estragole, Methyl cinnamate, Eugenol and Limonene and these components were detected in all samples in different locations. The previous components occupied about 78 % of the oil content. The sample location was very affective in changing the percentages of these constituents. Despite of

the location, the major component in all samples was Linalool which recorded 27.21, 26.57, 27.82 and 27.89 % for Tarabah, Raniah, Alhada and Alhaweiah, respectively. Estragole came in the second record and also affected by the previous locations.

Table (1): Volatile oil percentage and composition of sweet basil plant in different location in Taif Governorate

		Oil com							
Location	Oil (%)	1, 8-cineole	Linalool	Estragole	Methyl cinnamate	Eugenol	Limonene	Total*	
Tarabah	0.68	8.72	27.21	16.89	5.41	10.26	3.79	72.28	
Raniah	0.71	7.87	26.57	17.12	6.32	12.15	3.97	74.00	
Alhada	0.79	8.95	27.82	14.14	8.87	16.67	4.11	80.56	
Alhaweiah	0.78	9.13	27.89	18.78	5.96	14.98	3.15	79.89	

\*Total means, the total of the main components presented in the table not all the identified components. Although some components were identified they were not presented her because of their lower percentages.

## 3.2. Rosemary volatile oil

The volatile oil content of rosemary grown in different locations was showed in Table (2). The volatile oil percentage was affected by the sample location since it was 0.87, 0.92, 1.10 and 1.17% in Tarabah, Raniah, Alhada and Alhaweiah locations, respectively. Not only oil percentage but also oil composition was changed by growing rosemary in various locations. $\alpha$ -pinene, Camphine, 1,8-cineole, Verbinone and Borneol

were the main components of the oil. These five components were detected in the oil for all samples however; the percentages of the peaks were differed. Interestingly, the major component was differed also according to the growing area since it was 1,8-cineole in Tarabah location whereas; it was  $\alpha$ -pinene in the other sites of the experiment. Moreover, borneol resulted in the lowest values in oil composition in all samples

 Table (2): Volatile oil percentage and composition of rosemary plant in different location in Taif Governorate

Location	Oil (%)	Oil comp					
		α-pinene	Camphine	1,8-cineole	Verbinone	Borneol	Total*
Tarabah	0.87	22.57	4.97	29.11	7.85	3.11	67.61
Raniah	0.92	27.88	3.95	25.14	10.72	3.67	71.36
Alhada	1.10	34.78	4.13	27.42	11.25	8.34	85.92
Alhaweiah	1.17	29.55	4.22	24.88	11.54	6.65	76.84

\*Total means, the total of the main components presented in the table not all the identified components. Although some components were identified they were not presented her because of their lower percentages.

## 3.3. Marjoram volatile oil

It is evident from data tabulated in (Table 3) that the growing site was very important for marjoram volatile oil. Both volatile oil percentage and composition were affected by the location of samples. Samples from Alhada site recorded the highest volatile oil percentage (0.74 %) while the lowest value in this respect (0.61%) was obtained from samples collected from Raniah region. The GC-MS analysis of marjoram volatile oil showed that there were seven components recoded about 75% of oil content. These components were 1, 8cineole, Linalool, Methyl chavicol,  $\alpha$ -Terpinene,  $\gamma$ -Terpinene, Terpinene-4-ol and Sabinene.The main component in all locations was Linalool while Sabinene was the lowest component in this concern.

 Table (3): Volatile oil percentage and composition of marjoram plant in different location in Taif Governorate

	Oil (%)	Oil components (%)							
Location		1, 8-cineole	Linalool	Methyl chavicol	α- Terpinene	γ- Terpinene	Terpinene- 4-ol	Sabinene	Total*
Tarabah	0.63	3.37	31.27	19.24	3.37	5.27	13.27	2.27	78.06
Raniah	0.61	2.58	33.72	17.55	3.32	5.13	12.14	2.13	76.57
Alhada	0.74	4.87	37.15	13.72	7.75	6.82	14.78	2.88	87.97
Alhaweiah	0.71	7.12	32.64	13.78	6.88	6.76	13.68	2.64	83.50

\*Total means, the total of the main components presented in the table not all the identified components. Although some components were identified they were not presented her because of their lower percentages.

## **3.4. Rose volatile oil**

Our results concerning the volatile oil content of rose cultivated in different locations was shown in Table (4). Volatile oil was extracted from flowers grown at three locations which famous for rose production in Taif Governorate. The volatile oil percentage was slightly affected by the growing location since the percentages of volatile oil obtained from these locations were 0.033, 0.037 and 0.023 % for Al Shafa, Alhada and Alhaweiah, respectively. The GC-MS analysis of rose oil indicated that the main components of oil were  $\beta$ -linalool, Phenyl  $\alpha$ -Terpineol, Nerol, β-Citronellol, ethanol. Geraniol. Eugenol, Nonadecane and Heinocosane. In all growing locations, the major component of rose oil was Geraniol and the cultivation site had a little effect on this component under the environment of this study. β-Citronellol, Nerol and Nonadecane came in the second rank of oil components after Geraniol.

## 4. Discussion

Our results obtained in this study clearly indicate that the volatile oil percentage and its composition of basil, rosemary, marjoram and rose were affected by the different growing locations. As shown in Table (1) the growing location was an important factor for volatile oil variation in basil oil. The main components of basil oil confirmed the previous findings of <sup>[17, 30,</sup> <sup>36,72]</sup>. These results are in accordance with the others obtained by different authors in several There are usually considerable countries. variations in the content of the major components of basil obtained from different geographical origins <sup>[50]</sup>. The chemical composition of basil volatile oil was varied according to the plant growing location and some components were disappeared in some locations and presented in the others [76]. Not only the type of cultivar but also the agronomical practices and environmental conditions affect the composition of sensory important compounds <sup>[31, 70]</sup>. Regardless of these factors, 1,8-cineole, methyl cinnamate, methyl chavicol, and linalool <sup>[39]</sup> are generally the main compounds responsible for the typical basil aroma. Such variations in the essential oil content of basil across countries might be attributed to the varied agro climatic conditions of the regions <sup>[18, 72]</sup>. In the same direction, <sup>[21]</sup> reported similar trend.

Table 4: Volatile oil percentage and composition of rose plant in different location in Taif Governorate

		Oil components (%)									
Location	Oil (%)	β-linalool	Phenyl ethanol	α-Terpineol	Nerol	β-Citronellol	Geraniol	Eugenol	Nonadecane	Heinocosane	Total*
Al Shafa	0.033	8.22	3.54	4.67	11.72	14.64	20.62	3.55	11.36	6.22	84.54
Alhada	0.037	8.18	3.24	4.81	12.00	14.13	22.77	3.76	11.20	6.01	86.10
Alhaweiah	0.023	10.27	3.62	4.22	11.89	13.92	21.55	4.13	12.47	7.89	89.96

\*Total means, the total of the main components presented in the table not all the identified components. Although some components were identified they were not presented her because of their lower percentages.

Changing the growing location of rosemary led to a variation in volatile oil percentage as well as its composition (Table 2). The geographical region changed also the main component of oil since it was varied according to the sample site. In the same direction <sup>[28]</sup> reported that the camphor component was not detected in rosemary oil composition however different authors observed it <sup>[60, 74]</sup>.

The results of GC-MS analysis of rosemary volatile oil in our study identified the main components in all growing sites and these components were detected in all samples. The same components were previously detected in rosemary oil by several authors <sup>[28, 52, 60, 74]</sup>. Our results support the others obtained by <sup>[4]</sup> who reported that the volatile oil composition of rosemary may vary according to geographical and climatic regions because the oil contains mainly monoterpenes and sesquiterpenes which easily change bv environmental conditions and geographic origin.

In addition, different chemotypes of rosemary oil were identified based on main compounds of essential oils such as  $\alpha$ -pinene and verbenone <sup>[52]</sup> or 1,8-cineole, verbenone and camphor <sup>[16]</sup> according to geographical and climatic regions. The changes in volatile oil percentage of rosemary cultivated in different regions may be attributed to certain environmental factors that might stimulate the essential oil production by the

rosemary plants <sup>[33]</sup>. Different studies can be found in the scientific literature regarding rosemary essential oil yield and how it changes depending on the plant geographical origin. For example, <sup>[4]</sup> reported differences at the essential oil yield of the Sardinian R. officinalis L., collected from different natural stations. For these authors, yields obtained from the northern and eastern samples were, on average, 2-fold higher than those of the southern and central samples, although they did not relate these variations to any edaphoclimatic condition in particular. Later, <sup>[75]</sup> described the essential oil yielded by R. officinalis var. typicus and var. troglodytorum endemic to Tunisia, growing wild in different bio climates. In this case, the essential oil yield for the typicus variety was higher in upper semiarid zones than that obtained from sub-humid regions. From these results it is clear that rosemary essential oil is influenced by the habitat in which grow. the plants However. the main edaphoclimatic factor that modifies this parameter is not clear.

The climate of the zone plays an important role at the essential oil level production <sup>[34</sup>]. In the same variety, rosemary essential oil yield increases in bioclimatic zones with lower thermicity indexes <sup>[75]</sup>. Although, at a quantitative level, the essential oil chemical composition undergoes variations over the different bioclimatic areas studied, the relative concentration of the three components that define the rosemary essential oil chemotype (1,8-cineol, camphor and  $\alpha$ -pinene) did not show statistically significant differences between the zones studied. Thus, no differentiation related to a specific chemotype can be defined depending on the bioclimatic area. These results are in agreement with those published by <sup>[75]</sup>, who affirmed that variations in the chemical composition of rosemary essential oil from Tunisia should be attributed to varieties rather than bioclimatic conditions. On the other hand, the chemical composition of 32 rosemary samples collected at the same time from 12 different sites in the north of Algeria highlighted a strong correlation between the volatile chemical compositions of the samples and their origins <sup>[66]</sup>. The growing location was also affected the volatile oil content of marjoram (Table 3). As our results indicated it could be noticed that there were variations in volatile oil percentages and composition according to the site samples. It has been reported that the geographical region is one of the important factors affecting the composition of volatile oil in marjoram <sup>[5, 29, 42]</sup>. Moreover, the previous factors are interdependent and influence one another <sup>[65]</sup>. There are many reports in the literature showing the variation in the yield and chemical composition of the essential oil with respect to geographical regions [16, 61, 67, 68].

The other factor may affecting the oil composition of marjoram is altitude. There was previously findings support this idea <sup>[71]</sup>. In the same trend, <sup>[67]</sup> also reported that the essential oil yield and composition varied significantly, depending on the locations where the plants grew. Our results concerning rose oil content were tabulated in Table (4). From our data it could be noticed that the growing location was affected the oil percentage and its composition. The main components obtained from oil analysis support the others obtained previously by several authors. For example, <sup>[9]</sup> used gas chromatography/mass spectrometry (GC/MS) analysis for Taif rose oil and he found that citronellol, geraniol and 5methyl octadecane were reported to be the main constituents of oil. In addition, <sup>[69]</sup> illustrated that citronellol, geraniol, nerol, nonadecane and heneicosane to be the major components of the rose essential oil. Moreover, Rose oil samples were high in citronellol, geraniol and nerol <sup>[58, 38]</sup>. Therefore, our results suggest that the variations in volatile oil components are likely the result of environmental and processing differences and climatic conditions of Taif Governorate are suitable for the production of rose oil of international standards.

Generally, our results discussed here concluded that it will be useful to study the conditions of the location where aromatic plants will be cultivated since the geographical regions, even in the same country, affected not only the volatile oil percentage but also its composition. More studies in this area of research are recommended in order to explain exactly how the environmental conditions as well as altitude can affect the oil content of aromatic plants.

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Vol. 1 Issue. 5 2013

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