

Evaluation of the Chemical Profiling, Total Phenolic Composition, the Antioxidant and Antimicrobial Properties of the Essential Oils of *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L.

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Abstract: The purpose of this study was to determine the chemical composition, total phenolic composition, the antioxidant and antimicrobial properties of the essential oils (EOs) of *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L. The essential oils of *M. piperita* L., *S. officinalis* L., and *T. vulgaris* L. were analyzed by means of gas chromatography-mass spectrometry (GC-MS) to demonstrate their chemical composition. The antioxidant properties of the EOs were evaluated with the 2,2-diphenyl-picrylhydrazyl (DDPH) free radical scavenging assay, their total phenolic compound contents were determined by the Folin Ciocalteu method, and their antimicrobial activities were evaluated by the disc diffusion assay. The major compounds in the contents of the essential oils of *M. piperita* L., *S. officinalis* L., and *T. vulgaris* L. were found to be eucalyptol, 1R- α -pinene, and o-cymene, respectively. In the 2,2-diphenyl-picrylhydrazyl (DDPH) assay, the EO of *M. piperita* L. (8,930.01 μ MTE/g) demonstrated the highest antioxidant activity, followed by the activities of the EOs of *T. vulgaris* L. (157.76 μ MTE/g) and *S. officinalis* L. (115.54 μ MTE/g). The total phenolic compound contents of *M. piperita* L., *T. vulgaris* L., and *S. officinalis* L. were measured as 135.074, 0.242, and 0.221 mMGAE/g. All essential oils showed antioxidant activities and antimicrobial activities. The highest antimicrobial activity against *S. aureus*, *A. nigeris*, and *C. albicans* was determined in the EO of *M. piperita* L. within diameters of 42, 32, and 28 mm, respectively. These properties of essential oils are used in the pharmaceutical and food industries. The essential oils are approved as official medications in many countries and take place in their pharmacopoeias.

Key words: Essential oils, chemical composition, total phenolic composition, antioxidant capacity, antimicrobial activity.

1. Introduction

The plant life of Turkey, which is called the flora, consists of diverse species. The number of all plant taxa reaches figures over 12,000 and it is estimated that they belong to more than 9,000 species. Of these species, more than 33% are classified under the title “endemic species” and a similar portion is composed of aromatic plants [1].

Essential oils are incorporated as flavouring agents in several industrial products including food, beverages, spirits, perfumes, pharmaceuticals, and cosmetics [2]. A few aromatic and aliphatic

constituents are present in the composition of the EOs, however, the main compounds in the essential oils (EOs) are terpenoids and phenylpropanoids, with monoterpenes, sesquiterpenes, and their oxygenated derivatives composing the largest group of chemical entities [3]. The bioactivity of a particular EO is usually determined by either one or two of its main components, however, the overall activity is not always attributable to the individual molecules in the composition [4]. Rather, it is sometimes yielded as a result of the combination of the activities of the composing molecules or modification of their activities under the effect of several interactions.

Natural products and their derivatives are important sources of novel therapeutic molecules [5]. The

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essential oil composition of plants is highly influenced by the genetic and environmental factors, the age of organs, climate conditions, the features of the organs, seasons, and the culture site. Because of the vast multiplicity of the variations, the essential oil composition derived from a specific plant sometimes does not match the profile defined by Standard ISO 9909 [6].

Many plant genera with several well-recognized activities like chemotherapeutic, antiviral, antimicrobial, antimutagenic, antioxidant, and anti-inflammatory properties are members of the Lamiaceae family. Some of the products derived from this family are also beneficial when used for the relief of some intestinal disorders and some clinical conditions of bronchitis. *Mentha piperita* L., *Salvia officinalis* L. and *Thymus capitatus* L. are some of the popular examples of these types of plants from Lamiaceae family [7, 8]. The family Lamiaceae includes 252 genera and more than 6,700 species approximately [9].

Mentha piperita L., *Salvia officinalis* L., and *Thymus vulgaris* L. are aromatic plants in Lamiaceae family and the products derived from these plants are used as medications or spice almost widely worldwide [10]. The non-medicinal use of thyme is worthy of attention because thyme is used in the food and aroma industries; it is widely used as a culinary ingredient and it serves as a preservative for foods especially because of its antioxidant effect. The EO of thyme constitutes the raw material in perfumery and cosmetics due to its special and characteristic aroma [11]. The EOs derived from *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L. are mainly used to relieve the cough, cold, digestive issues, menstrual symptoms, pain, headaches, skin problems, and to resolve obstructed sinuses [12].

This study aimed to extract the EOs from *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L. to identify the constituents in their composition using the gas chromatography-mass spectrometry

(GC-MS) analysis, to evaluate their antimicrobial and antioxidant activities, and to determine their total phenolic compounds in their composition.

2. Materials and Methods

2.1 Plant Material

The aerial parts from *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L. were randomly collected from Adana, Turkey. Plant materials were harvested during the flowering period between March 2018 and April 2018. The taxonomic identification of plant materials was confirmed by a plant taxonomist, Dr. Hikmet Ozbek, Professor in Entomology, retired from the Department of Plant Protection, Ataturk University, Erzurum, Turkey. While the plant leaves were still on their stems, they were dried at the room temperature at a shadowy environment during a period of 6 days. After this period, the plant leaves were separated from their stems.

2.2 Essential Oil Extraction Process

The aerial parts of the air-dried plants of *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L. were subjected to the extraction process in a modified Clevenger-type apparatus for 4 h. After deriving the EOs, they were exposed to a drying process with anhydrous sodium sulfate (Na_2SO_4) [13]. Then, they were deoxygenated by being exposed to gaseous nitrogen and stored in a sealed vial at -4°C until the time of analysis. Qualitative analyses of the EOs were performed using GC/MS.

2.3 Test Organisms

The antimicrobial activity of the derived EOs was tested against five microorganism species, namely *Staphylococcus aureus* (gram-positive bacteria), *Escherichia coli* (gram-negative bacteria), *Pseudomonas aeruginosa* (gram-negative bacteria), *Aspergillus niger* (fungus), and *Candida albicans* (yeast).

2.4 Determination of the Antimicrobial Activities of the Essential Oils

The antimicrobial activities of the EOs were evaluated by disc diffusion assay. Discs of 6 mm in diameter made of Whatman No. 1 filter paper were used. Briefly, a 100 μ L suspension from the individual test microorganism was spread homogeneously on each plate of mannitol salt agar media. Each disc was soaked with 100 μ L of EOs diluted at a rate of 10% and then were placed on the microbial lawn. To perform the positive control experiments, the same processes were repeated but instead of using the respective EOs, ofloxacin was used to test the antibacterial activity and nystatin for antifungal activity. The negative control experiments were carried out similarly repeating the same processes but by using sterile water instead of using the EOs [14]. The tests were repeated three times to ensure reliability. All plates were then incubated at 37 °C for 24 h. The emergent antibacterial halos were measured in millimeters.

2.5 Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

The analysis of the essential oil was performed using a Thermo Finnigan Trace GC/TraceDSQ/A1300 equipped with an SGE-BPX5 MS capillary column (30 m \times 0.25 mm id, 0.25 μ m). For GC/MS detection an electron ionization system with an ionizing energy of 70 eV was used. The components were identified by comparing the respective retention times together with the mass spectra against the standards of Wiley 7N library data of the GC/MS system and against the data in the literature [15]. Furthermore, the results were also confirmed by comparing the elution order of the compounds with their relative retention indices on non-polar phases according to the reports in the literature [15].

3. The Evaluation of the Antioxidant Activities of the Essential Oils

3.1 2,2-Diphenyl-1-Picrylhydrazyl (DPPH) Free Radical Scavenging Assay

The antioxidant activities of the EOs were evaluated by the 2,2-diphenyl-picrylhydrazyl (DPPH) free radical scavenging assay, as described in the literature previously [16]. Briefly, a 10 μ L volume of each respective EO was diluted with methanol (HPLC-grade) to reach a final volume of 150 μ L. Then, 4 mg of DPPH was diluted in a 100 mL volume of methanol to obtain a working solution with an absorbance at 515 nm. Diluted EO solutions were then mixed with a 2.85 mL of DPPH and incubated for 24 h at room temperature in a dark environment. Finally, the absorbances at 515 nm were measured in a visible light spectrophotometer. Methanol (HPLC-grade) was used as the blank, and trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid, Sigma) was used as the standard curve. The antioxidant activity was expressed as μ g of trolox equivalents per 1 mL of EOs (μ g trolox equiv./g DE) [17]. All determinations were performed in triplicates.

3.2 Total Phenolic Content

Total phenolic compound contents were determined by the Folin Ciocalteu method [18]. The samples of the extracts (0.5 mL, 1:10 dilution) were mixed with Folin Ciocalteu reagent (5 mL, diluted at 1:10 with distilled water) for 5 min. Then, aqueous Na₂CO₃ (4 mL, 1 M) was added. The mixture was allowed to stand for 15 min and the phenols were determined by the colorimetric method at 765 nm. The standard curve was prepared using the standard solution of Gallic acid in methanol in the range of 20-200 μ g/mL ($R^2 = 0.987$) as a common reference compound. Total phenolic contents can be calculated from the formula: $T = CV/M$, where, T = Total Phenolic concentration,

C = Concentration of gallic acid from the calibration curve ($\mu\text{g/mL}$), V = Volume of extract (mL), $M = W_t$ of essential oils of plants [19].

4. Result and Discussion

The extracted essential oils obtained from the areal parts of *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L. were hydrodistilled for 4 h using a Clevenger apparatus. Next, the essential oils were qualitatively analyzed with GC-MS. Eucalyptol (cineole), cyclohexanone (5-methyl-2-(1-methyl ethyl)-cis), cyclohexanol(1-methyl-4-(1-methyl ethyl), and pulegone (cyclohexanone,5-methyl-2-(1-methyl ethylidene)-R) were the major compounds of *Mentha piperita* L. essential oil, however, trifluoroacetyl- α -terpineol, isopulegol, 2-acetylcyclohexanone, L-menthone, 4-isopropyl-1,3-cyclohexanedione, menthol, 3,7-dimethyl-7-octen-2-ol, citronellyl butyrate, menthyl acetate, and citronellol were found in reasonable amounts. The major chemical components are given in Table 1.

The EO of *M. piperita* L. was found to be rich in eucalyptol. Eucalyptol is monoterpene that is used for several purposes, including its use in medications, as well as its use as a flavoring and fragranting agent [20].

The EOs of *Mentha piperita* L. from Brazil include menthyl acetate, menthol, menthofuran, menthone, and 1,8 cineole are the major components [21]. On the other hand, the EOs extracted in different regions in Iran are composed of menthol, menthofuran, menthyl acetate, and 1,8-cineole as the major components, comprising 99.97% of their whole content [22, 23]. However, other results from Iran have shown that 93.58% of the total oil content includes α -terpinene, piperitone oxide, trans-carveol, and isomenthone as the main constituents. These results are similar to the results of the present study. However, the results from Brazil are different as presented above. Furthermore, the results from Colombia are different from the

results reported from Iran and from the results of this present study in the sense that the EOs from Columbia contain pulegone, isomenthol, and isomenthone as their major components in 99.43% of the EO content [24]. It is due to the fact that the major components of EOs may show variations depending on the geographical conditions, climate, and the effect of sunlight. Table 1 presents the results of the GC-MS analysis for the EOs derived from *Salvia officinalis* L.

The major components of the EO of *Salvia officinalis* L. were α -pinene, camphor, camphene, eucalyptol, α -linalool, 3-thujanone, bornanone, and α -terpineol. This composition is similar to those reported for the EOs derived from Italy, Romania, Serbia and Montenegro, Brazil and Turkey [25]. The essential oils analysed in this present study may offer a favorable potential for the treatment of coughs and unspecific irritations of the respiratory tract as an efficient agent against several foodborne pathogens, including yeasts, moulds, and gram positive bacteriae mainly due to camphor. The analyzed EOs contained large proportions of oxygenated components, represented by oxygenated monoterpenes, oxygenated sesquiterpenes, and oxygenated diterpenes [26]. The geographical origin, ecological conditions, and the genetic factors may be responsible for the high intraspecific variability observed in the essential oils of *S. officinalis*. In fact, these factors influence the plant's biosynthetic pathways, consequently resulting in the relative proportions of the main characteristic compounds in their composition [26].

Table 1 presents the results of the GC-MS analysis for the *Thymus vulgaris* L.

p -cymene, terpinene, thymol, carvacrol, and caryophyllene were the major compounds of the EO of *Thymus vulgaris* L., whereas, o -isopropylanisole, p -cymene-7 ol, 2,3,5-trimethylanisole, 3,4-diethylphenol, and 4-tert-butylphenol were found in its content at reasonable amounts. The volatile components are important in determining the biological activity of *Thymus* species. The essential oil

Table 1 The chemical composition (major compounds only) of the essential oils.

Essential oils	No.	Major compound of essential oils	Molecular formula	Molecular weight (g/mol)
<i>Mentha piperita</i> L.	1	Eucalyptol (cineole)	C ₁₀ H ₁₈ O	154
	2	Cyclohexanone(5-methyl-2-(1-methylethyl)-cis-)	C ₁₀ H ₁₈ O	154
	3	Cyclohexanol, 1-methyl-4-(1-methylethyl)	C ₁₀ H ₂₀ O	156
	4	Pulegone(cyclohexanone,5-methyl-2-(1-methylethylidene)-R)	C ₁₀ H ₁₆ O	152
	5	Trifluoroacetyl- α -terpineol	C ₁₂ H ₁₇ F ₃ O ₂	250
	6	Isopulegol	C ₁₀ H ₁₈ O	154
	7	2-acetylcyclohexanone	CH ₃ COC ₆ H ₉ (=O)	140
	8	L-menthone	C ₁₀ H ₁₈ O	154
	9	4-isopropyl-1,3-cyclohexanedione	C ₈ H ₁₄ O ₂	154
	10	Menthol	C ₁₀ H ₂₀ O	156
	11	3,7-dimethyl-7-octen-2-ol	C ₁₀ H ₂₀ O	156
	12	Citronellyl butyrate	C ₁₄ H ₂₆ O ₂	226
	13	Menthyl acetate	C ₁₂ H ₂₂ O ₂	198
	14	citronellol	C ₁₀ H ₂₀ O	156
<i>Salvia officinalis</i> L.	1	1R- α -pinene((1R)-2,6,6-trimethylbicyclo(3.1.1)hept-2-ene)	C ₁₀ H ₁₆	136
	2	Camphenen(bicycle(2.2.1)heptanes,2,2-methyl-3-methylenen-)	C ₁₀ H ₁₆	136
	3	2(10)-pinene(bicycle(3.1.1)heptanes,6,6-dimethyl-2-methylene)-(1S)	C ₁₀ H ₁₆	136
	4	Eucalyptol(cineole)	C ₁₀ H ₁₈ O	154
	5	α -linalool(1,6-octadien-3-ol, 3,7-dimethyl-)	C ₁₀ H ₁₈ O	154
	6	3-thujanone(bicycle(3.1.0)hexan-3-one,4-methyl-1-(1-methylethyl)-	C ₁₀ H ₁₆ O	152
	7	(+)-2-bornanone(bicycle(2.2.1)heptan-2-one,1,7,7-trimethyl-(1R)	C ₁₀ H ₁₆ O	152
	8	α -terpineol(3-cyclohexene-1-methanol, α 4-trimethyl-)	C ₁₀ H ₁₆ O	154
<i>Thymus vulgaris</i> L.	1	O-cymene(benzene,1-methyl-4-(1-methylethyl)-)	C ₁₀ H ₁₄	134
	2	Terpinene(1,4-cyclohexadiene,1-methyl-4-(1-methylethyl)-)	C ₁₀ H ₁₆	136
	3	Thymol(phenol,5-methyl-2-(1-methylethyl)-)	C ₁₀ H ₁₄ O	150
	4	Carvacrol(phenol,2-methyl-5-(1-methylethyl)-)	C ₁₀ H ₁₄ O	150
	5	Caryophyllene(bicycle(7.2.0)undec-4-ene,4,11,11-trimethyl-8-methylene-)	C ₁₅ H ₂₄	204
	6	o-isopropylanisole	C ₁₀ H ₁₄ O	150
	7	p-cymen-7-ol	C ₁₀ H ₁₄ O	150
	8	2,3,5-trimethylanisole	C ₁₀ H ₁₄ O	150
	9	3,4-diethylphenol	C ₈ H ₁₀ O	122

content and quantitative composition of *Thymus vulgaris* L. can be influenced by the harvest time, and by the ecological and climatic conditions. A correlation has been reported between the soil type and the chemotype of the *Thymus vulgaris* L. [14]. Thymol is the major component of *Thymus vulgaris* L. according to the analysis results from Morocco, Iran, and Algeria [26]. In most of the studied cases, there is a linear relationship between the thymol concentrations and carvacrol. If thymol is in high concentrations in the EO, carvacrol amount is found to be low and the reverse holds true as well [14, 5].

The DPPH free radical scavenging activities and total phenolic contents in the EOs of *Mentha piperita* L., *Salvia officinalis* L. and *Thymus vulgaris* L. are shown in Table 2.

The antioxidant activity of the three types of EOs in this study was measured by the DPPH assay. Their activities ranged from 115.54 to 8,930.01 μ M Trolox equiv./DE. Among the investigated essential oils, the EO of *Mentha piperita* L. had the highest free radical scavenging activity at 8,930.01 μ M Trolox equiv./DE. This activity was followed by that of *T. vulgaris* (157.76 μ M Trolox equiv./DE) and *S. officinalis*

Table 2 Antioxidant activity and total phenolic compounds of essential oils.

Essential oils	Antioxidan activity (DPPH) μ MTE/g	Phenolic compound (Folin) mMGAEG/g
<i>Mentha piperita</i> L.	8,930.01	135.074
<i>Salvia officinalis</i> L.	115.54	0.242
<i>Thymus vulgaris</i> L.	157.76	0.221

(115.54 μ M Trolox equiv./DE). The high scavenging activity of *Mentha piperita* L. can be explained partially by the presence of a higher amount of compounds responsible for this activity, including cineole, cyclohexanone, cyclohexanol, pulegone, and sesquiterpene hydrocarbons, revealed in the analysis of the respective EO [27].

The phenols were expressed in terms of gallic acid equivalent (GAE) per millimolar of dry weight basis. The results revealed that total phenolic content varied among different plant EOs. The total phenolic content was found to be highest in the EOs extracted from *Mentha piperita* L. (135.074 mM GAE/g DE) compared to *Thymus vulgaris* L. (0.242 mM GAE/g DE) and *Salvia officinalis* L. (0.221 mM GAE/g DE). The earlier studies reported that the antioxidant activity of sage was 346.61 μ M TE/mg [26].

The high DPPH radical scavenging ability of the EOs might result from the high phenolic content. In the previous studies investigating the antioxidant activity of the herbal EOs, it was reported that the antioxidant activity of many EOs was directly proportional to the concentration of the phenolic compounds they contained. Therefore, a causative association between these two variables was suggested [28]. The main compounds of the EO derived from *T. vulgaris* L. were the natural terpenoid thymol and its phenol isomer carvacrol, which had antioxidative, antimicrobial, and antibacterial effects. Therefore, it can be concluded that the essential oil of *T. vulgaris* has a potential antioxidant activity and a protective effect against the toxicity of aflatoxins, and this protective effect is dependent [29].

The antimicrobial activities of the EOs were tested against three bacterial strains, one fungus, and one yeast, using the disc diffusion method. The results of

the antimicrobial tests are presented in Table 3.

The antibacterial activity was highest among all essential oils. The EO of *Mentha piperita* L. exhibited a significant antimicrobial activity against *Staphylococcus aureus* (42 mm), *Escherichia coli* (24 mm), and *Pseudomonas aeruginosa* (10 mm). On the other hand, the EOs of *Salvia officinalis* and *Thymus vulgaris* showed narrower inhibition zones against *Staphylococcus aureus* (8 mm), *Escherichia coli* (9 mm), and *Pseudomonas aeruginosa* (8 mm). The antifungal activities of the EOs are presented in Table 3. The essential oils showed strong antifungal activity against yeast and fungi strains. *Mentha piperita* L. showed strong antifungal activity against *A. nigeris* (32 mm) and *C. albicans* (28 mm). *Salvia officinalis* and *Thymus vulgaris* exhibited a moderate antifungal activity against *A. nigeris* and *C. albicans* 7 mm and 8 mm, respectively. Compared to ofloxacin, the essential oil of *M. piperita* L. showed a stronger antibacterial activity especially against *S. aureus*, *E. coli*, and *P. aeruginosa*. Compared to nystatin, the essential oils of *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L. also appeared to be more active against *N. nigeris* and *C. albicans*. The antimicrobial properties of the essential oils are assumed to be associated with the oxygenated compounds in their composition. However, the amounts of the oxygenated components were smaller in these three types of EOs in this present study, but the administration of these EOs resulted in a moderate antimicrobial activity, disproportionally to their compositions [30]. The present study reveals that the EOs extracted from the aerial parts of *Mentha piperita* L., *Salvia officinalis* L., and *Thymus vulgaris* L. offer antibacterial and antifungal potentials. These results may be promising for antibacterial and antifungal

Table 3 Antimicrobial activity of essential oils.

Bacterial strains	Gram-positive	Gram negative		Fungus	Yeast
Essential oils	<i>Staphylococcus aureus</i> (ATCC 6338)	<i>Escherichia coli</i> (ATCC 10536)	<i>Pseudomonas aeruginosa</i> (ATCC 15442)	<i>Aspergillus nigeris</i>	<i>Candida albicans</i>
<i>Mentha piperita</i> L.	42 mm	24 mm	10 mm	32 mm	28 mm
<i>Salvia officinalis</i> L.	8 mm	9 mm	8 mm	7 mm	8 mm
<i>Thymus vulgaris</i> L.	8 mm	9 mm	8 mm	7 mm	7 mm
Ofloxacin	22 mm	26 mm	20 mm		
Nystatine				18 mm	20 mm

activities against a wider range of microorganisms. These results are important considering the multiple antibiotic resistance of the bacteria [14].

Some researchers reported a relationship between the chemical structures of the most abundant compounds in the essential oils and their antimicrobial or antifungal activities. This might explain the fact that the synergistic or antagonistic effect of one compound in the EO content has to be taken into account even though it exists in a minor percentage in the composition of the mixture. This information in the literature should be remembered, too, although the antimicrobial activity of the EOs is attributed mainly to their main compounds in their contents [31].

These properties of essential oils are used in the pharmaceutical and food industries. The essential oils are approved as official medications in many countries and take place in their pharmacopoeias.

References

- [1] Baser, K. H. C., and Buchbauer, G. 2015. *Handbook of Essential Oils: Science, Technology, and Applications* (2nd ed.). CRC Press Reference.
- [2] Sarkic, A., and Stappen, I. 2018. "Essential Oils and Their Single Compounds in Cosmetics—A Critical Review." *Cosmetics* 5: 11.
- [3] Dhifi, W., Bellili, S., Jazi, S., Bahloul, N., and Mnif, W. 2016. "Essential Oils Chemical Characterization and Investigation of Some Biological Activities: A Critical Review." *Medicines* 3 (4): 25.
- [4] Altemimi, A., Lakhssassi, N., Baharlouei, A., and Watson, D. G. 2017. "Phytochemicals: Extraction, Isolation, and Identification of Bioactive Compounds from Plant Extract." *Plants* 6: 42.
- [5] Rasoanaivo, P., Wright, C. W., Willcon, M. L., Gilbert, B. 2011. "Whole Plant Extracts versus Single Compounds for the Treatment of Malaria: Synergy and Positive Interactions." *Malaria Journal* 10 (1).
- [6] Bernotiene, G., Nivinskiene, O., Butkiene, R., and Mockute, D. 2007. "Essential Oil Composition Variability in Sage (*Salvia officinalis* L.)." *Chemija* 18 (4): 38-43.
- [7] Tzima, K., Brunton, N. P., and Rai, D. K. 2018. "Qualitative and Quantitative Analysis of Polyphenols in Lamiaceae Plants—A Review." *Plants (Basel)* 7 (2): 25.
- [8] Hussain, A. I., Anwar, F., Iqbal, T., and Bhatti, I. A. 2011. "Antioxidant Attributes of Four Lamiaceae Essential Oils." *Pak J Bot* 43 (2): 1315-21.
- [9] Hedge, C. 1992. "A Global Survey of the Biogeography of the Labiatae." In *Advances in Labiatae Science*, edited by Harley RM & Reynolds T. Royal Botanic Gardens, Kew, 7-17.
- [10] Samarth, R. M., Samarth, M., and Matsumoto, Y. 2017. "Medicinally Important Aromatic Plants with Radioprotective Activity." *Future Science* 3 (4).
- [11] Luaibi, N. M., Jasim, M. G., and Mousa, A. H. 2017. "A Study Medicinal and Nutritional Effects of Thyme on Hematological Changes in Male Albino Rats." *Iraqi Journal of Science* 58 (3): 1177-88.
- [12] Ali, B., Al-Wabel, N. A., Shams, S., Ahamad, A., Khan, S. A., and Anwar, F. 2015. "Essential Oils Used in Aromatherapy: A Systemic Review." *Asian pacific Journal of Tropical Biomedicine*.
- [13] Demir, H., and Kalayci, S. 2017. "Chemical Composition and Antimicrobial Activity of Essential Oils of *Ocimum basilicum* var. album (L.) Benth, *Lavandula angustifolia* subsp. *Angustifolia*, *Melissa officinalis* Belonging to Lamiaceae Family." *Journal of Food Science and Engineering* 7: 461-71.
- [14] Nezhadali, A., Masromia, M., Solati, A., Akbarpour, M., and Nakai, M. M. 2009. "Analysis of the Flower Essential Oil at Different Stage of Plant Growth and *in Vitro* Antibacterial Activity of *Perovskia abrotanoides* Karal in Iran." *Der Pharma Chem* 1: 146-50.
- [15] Gulluce, M., Sokmen, M., Sahin, F., Sokmen, A., Adiguzel, A., and Ozer, H. 2004. "Biological Activities of the Essential Oil and Methanolic Extract of *Micromeria fruticosa* (L.) Druce ssp. *Serpyllifolia* (Bieb) pH Davis Plants from the Eastern Anatolia Region of

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- Turkey." *J Science of Food and Agriculture* 84: 735-41.
- [16] Scherer, R., and Godoy, H. T. 2009. "Antioxidant Activity Index (AAI) by the 2,2-Diphenyl-1-Picrylhydrazyl Method." *Food Chem.* 112: 654-8.
- [17] Julia, M. M., Luciane, B., and Balbina, A. A. 2018. "Chemical Characterization of Polyphenol Extracts from Andean and Industrial *Solanum tuberosum* Tubers and Their Cytotoxic Activity on Human Hepatocarcinoma Cells." *SDRP Journal of Food Science Technology* 2 (2): 1-13.
- [18] Blainski, A., Lopes, G. C., and Plazzo de Mello, J. C. 2013. "Application and Analysis of the Folin Ciocalteu Method for the Determination of the Total Phenolic Content from *Limonium brasiliense* L." *Molecules* 18: 6852-65.
- [19] Asadi, S., Ahmadiani, A., Esmaeili, M. A., Sonboli, A., Ansari, N., and Khodaghali, F. 2010. "In Vitro Antioxidant Activities and an Investigation of Neuroprotection by Six *Salvia* Species from Iran: A Comparative Study." *Food and Chemical Toxicology* 48: 1341-9.
- [20] Luis, A., Duarte, A. P., Pereira, L., and Domingues, F. 2017. "Chemical Profiling and Evaluation of Antioxidant and Antimicrobial Properties of Selected Commercial Essential Oils: A Comparative Study." *Medicines* 4 (36).
- [21] Scavroni, J., Boaro, C. S. F., Marques, M. O. M., and Ferreira, L. C. 2005. "Yield and Composition of the Essential Oil of *Mentha piperita* L. (Lamiaceae) Grown with Biosolid." *Braz. J. Plant Physiol* 17: 345-52.
- [22] Moghaddam, M., Pourbaige, M., Tabar, H. K., Farhadi, N., and Hosseini, S. M. A. H. 2013. "Composition and Antifungal Activity of Peppermint (*Mentha piperita*) Essential Oil from Iran." *Journal of Essential Oil Bearing Plants* 16 (4).
- [23] Davod, Y., Gachkar, L., Rezaei, M. B., Taghizadeh, M., Astaneh, S. A., and Rasooli, I. 2006. "Biochemical Activities of Iranian *Mentha piperita* L. and *Myrtus communis* L. Essential Oils." *Phytochemistry* 67: 1249-55.
- [24] Reddy, D., Al-Rajab, A. J., Sharma, M., and Albratty, M. 2017. "Chemical Constituents, in Vitro Antibacterial and Antifungal Activity of *Mentha piperita* L. (Peppermint) Essential Oils." *Journal of King Saud University-Science.*
- [25] Craft, J. D., Satyal, P., and Setzer, W. N. 2017. "The Chemotaxonomy of Common Sage (*Salvia officinalis*) Based on the Volatile Constituents." *Medicines* 4 (47).
- [26] Farhat, M. B., Jordan, M., Chaouech-Hamada, R., Landoulsi, A., and Sotomayor, J. A. 2009. "Variations in Essential Oil, Phenolic Compounds, and Antioxidant Activity of Tunisian Cultivated *Salvia officinalis* L." *Journal of Agricultural and Food Chemistry* 57: 10349-56.
- [27] Grulova, D., De Martino, L., and Mancini E. 2015. "Seasonal Variability of the Main Components in Essential Oil of *Mentha × piperita* L." *J Sci Food Agric* 95 (3): 621-7.
- [28] Grigore, A., Paraschiv, I., Colceru-Mihul, S. C., Bubueanu, C., Draghici, E., and Ichim, M. 2010. "Chemical Composition and Antioxidant Activity of *Thymus vulgaris* L. Volatile Oil Obtained by Two Different Methods." *Romanian Biotechnological Letters* 15 (4): 5436-43.
- [29] El-Nekeety, A. A., Mohamed, S. R., Hathout, A. S., Hassan, N. S., Aly, S. E., and Abdel-Wahhab, M. A. 2011. "Antioxidant Properties of Thymus Vulgaris Oil Against Aflatoxin-Induced Oxidative Stress in Male Rats." *Toxicon* 57: 984-91.
- [30] Salleh, W. M. N. H. W., Ahmad, F., and Khong, H. Y. 2016. "Essential Oil Composition and Antimicrobial Activity of Piper Arborescents Roxb." *Marmara Pharmaceutical Journal* 20: 111-5.
- [31] Nieto, G. 2017. "Biological Activities of Three Essential Oils of the Lamiaceae Family." *Medicines* 4: 63.