Sema Carikci, Ahmet C. Goren*, Tuncay Dirmenci, Burhanettin Yalcinkaya, Aysen Erkucuk and Gulacti Topcu

Composition of the essential oil of *Satureja metastasiantha*: a new species for the flora of Turkey

https://doi.org/10.1515/znc-2019-0208
Received November 12, 2019; revised January 24, 2020; accepted January 30, 2020

Abstract: The aerial parts of *Satureja metastasiantha* were hydrodistilled for 3 h using a Clevenger-type apparatus. The essential oils were analyzed by gas chromatography/flame ionization detector and gas chromatography/mass spectrometry, simultaneously, the main compounds of which were characterized as *p*-cymene (22.3%), thymol (21.0%), carvacrol (18.4%), and γ-terpinene (12.1%). Antioxidant capacity, acetylcholinesterase and butyrylcholinesterase inhibition effects, and antimicrobial and antifungal properties of the species were evaluated. The anticholinesterase activity of the essential oil of *S. metastasiantha* was observed with 30% inhibition at 200 μg/mL. The essential oil of the species showed activity against *Staphylococcus aureus* with 128 μg/mL minimum inhibitory concentration value.

Keywords: AChE; antimicrobial; antioxidant; carvacrol; essential oil; *Satureja metastasiantha*.

1 Introduction

Species of Lamiaceae family, in the world and Turkey, has been used in a wide range of applications. Its members are widely used as a spice in Turkish folklore in flavoring condiments, relishes, soups, sausages, canned meats, and spicy table sauces [1, 2]. Lamiaceae is the third largest family based on its number of taxon and the fourth largest family based on its number of species in Turkey. The rate of endemism is around 44% [3]. The genus *Satureja* L. is represented by 46 and 16 species in the world and Turkey, respectively, 6 species of which in Turkey are endemic. The species of the genus *Satureja* are distributed mainly in Mediterranean countries, Europe, North Africa, Morocco, Saudi Arabia, Turkey, and Caucasus [4, 5], among which Turkey has the most of the species of genus *Satureja* [6, 7]. According to the recent studies on Lamiaceae family in Turkey, the number of species is on the increase [3, 8]. *Satureja metastasiantha* (Zap Kaya kekiği) is one of the species in the flora of Turkey, discovered by Dirmenci and his group [7]. *Satureja metastasiantha* is distributed in north of Iraq and Southeastern Anatolia of Turkey [7, 9].

Satureja species are popularly named as "Sivri Kekik" or "Kaya Kekiği" by local people in Turkey, and their species have commercial importance. Mainly, the six species of the genus, e.g. *Satureja thymbra*, *S. cuneifolia*, *S. wiedemanniana*, *S. spicigera*, *S. hortensis*, and *S. cilicia*, are used as spice (thyme), and their essential oils are important input in pharmaceuticals and cosmetic industries because of their antibacterial and antifungal activities. These species are traditionally used against cold and bronchitis and exported across the world in high quantities as thyme and essential oil [1, 10, 11].

In our previous reports, *Satureja* species were indicated to grow mainly in Western and Southern Anatolia because of the climate conditions [1]. The title species has been reported to be available in the Hakkari province of Turkey, having the most continental climate zone. *Satureja metastasiantha* is determined as the second species grown in strong continental climate zone of Turkey. In this study, the chemical composition of essential oil of *S. metastasiantha* is reported for the first time as a new source of commercially important species. The main components were found to be thymol, carvacrol, *p*-cymene, and γ-terpinene. Biological activities of the essential oil and methanol extract of the species are also reported.

***Corresponding author: Ahmet C. Goren,** Bezmialem Vakıf University, Faculty of Pharmacy, 34093 Fatih, Istanbul, Turkey, E-mail: ahmet.goren@acgpubs.org. https://orcid.org/0000-0002-5470-130X
Sema Carikci: Izmir Democracy University, Department of Environment Protection Technologies, Karabağlar, İzmir, Turkey. https://orcid.org/0000-0003-3657-9926
Tuncay Dirmenci: Department of Biology Education, Necatibey Faculty of Education, Balıkesir University, 10100 Balıkesir, Turkey
Burhanettin Yalcinkaya: TÜBİTAK UME, Chemistry Group Laboratories, 41400 Gebze-Kocaeli, Turkey. https://orcid.org/0000-0002-3744-6634
Aysen Erkucuk and Gulacti Topcu: Bezmialem Vakıf University, Faculty of Pharmacy, 34093 Fatih, Istanbul, Turkey

2 Materials and methods

2.1 Plant material

Aerial parts of *Satureja metastasiantha* Rech.f. were collected from 31 km of Şemdinli to Derecik, above Örencik village, K 37.163397 D 44.42670, 1600 m, on March 8, 2018. The plant was identified by Prof. Dr. Tuncay Dirmenci of Balikesir University, Turkey. A voucher specimen was deposited in the Herbarium of Department of Biology, Faculty of Necatibey Education Faculty, Balikesir University (TD 5126).

2.2 Isolation of the essential oils

The dried aerial parts (150 g × 3) of *S. metastasiantha* were subjected to hydrodistillation for 3 h, using a Clevenger-type apparatus. The obtained essential oil was dried over anhydrous $CaCl_2$ and stored at 4 °C until the measurement. The yields were determined as 0.80% ± 0.05. The density of the essential oil was measured to be 0.892 ± 0.002 g/mL.

2.3 GC and GC-MS conditions

The essential oil was analyzed by capillary gas chromatography (GC) and gas chromatography/mass spectrometry (GC-MS) systems.

2.3.1 GC-MS analysis

The essential oils were diluted with CH_2Cl_2 (1:10) and analyzed using a Trace 1310 GC series gas chromatograph (Thermo Scientific, Inc., Waltham, MA, USA) and a Thermo TSQ 9000 mass spectrometer (Thermo Scientific, Waltham, MA, USA). The separation was carried out in a DB5-fused silica column (60 m, 0.25 mm, ø with 0.5 µm film thickness). Helium was used as a carrier gas at a flow rate of 1 mL/min. The oven temperature was kept at 100 °C for 5 min, programmed to 240 °C at a rate of 4 °C/min and kept constant at 240 °C for 5 min. The injection and source temperatures were 250 °C and 220 °C, respectively. The MS interface temperature was 240 °C. The injection volume was 0.5 µL with a split ratio of 1:20. Injections of the samples were performed in triplicate. Electron ionization/MS was recorded at 70 eV ionization energy. The scan time was 0.5 s with 0.1 interscan delay. The mass range was from *m/z* 50 to 650 amu [1, 12].

2.3.2 GC analysis

The GC analysis was carried out using a Trace 1310 GC series gas chromatograph (Thermo Fischer Scientific, Inc., ON, CA). In order to obtain the same elution order with GC-MS, simultaneous injection was conducted by using the same column and an appropriate operational condition. FID temperature was set to 300 °C [12].

2.4 Identification of compounds

A homologous series of *n*-alkanes was used as a reference in the calculation of Kovats Indices (KI). Identification of the compounds was based on the comparison of their retention times and mass spectra with those obtained from authentic samples and/or the NIST and Wiley spectra, Library of Bezmialem Vakif University, Drug Application and Research Center (İLMER) as well as the literature data. Ultra Kit WRK 105 terpene mixture was used in co-injection to check the results of the library search. In addition, authentic samples for thymol, carvacrol, spathulenol, and caryophyllene oxide, previously isolated by our group from plant species, were used for identification of composition of the essential oil. The relative standard deviations (RSD%) of the identified compounds were calculated through three individual replicates of the obtained essential oil and determined to be 0.05%.

2.5 Antioxidant activity

The antioxidant activities of the essential oil and methanol extract were determined by using 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging and cupric (Cu^{2+}) ion reducing power assay (CUPRAC) methods [13, 14]. The results of the experiments are the average of triplicate analyses. The experimental data were calculated as the mean ± standard deviation. Variance ANOVA including one-way analysis was realized. Significant differences between the means were recorded by Duncan's multiple range tests. $p < 0.05$ was regarded as significant, and $p < 0.01$ was very significant.

2.5.1 DPPH free radical scavenging activity assay

The free radical scavenging activity of the essential oil and methanol extract were determined by DPPH free radical scavenging activity assay. Briefly, 0.1 mM solution of DPPH in ethanol was prepared, and 160 µL of this solution was

added to 40 μL of the sample solutions in ethanol (10, 25, 50, and 100 μM), which were then incubated for 30 min. As control standards, butylated hydroxytoluene (BHT), butylated hydroxyanisole (BHA), and α-tocopherol (α-TOC) were used. After the incubation at room temperature, the absorbance was measured at 517 nm.

2.5.2 Cu^{2+} reduction capacity (CUPRAC method)

For the determination of antioxidant capacity of the essential oil and methanol extract of species CUPRAC (Cupric Reducing Antioxidant Capacity), the following method was applied: Briefly, 1 M NH_4Ac (pH = 7.0), 10 mM $CuCl_2$, 7.5 mM Neocuproine, 1 M NH_4CH_3COO (pH 7.0), and distilled water were mixed in volume ratios of 1:1:1:0.6. Of the mixture, 180 μL was aliquoted into the wells. Then, 25 μL of the aliquot from the 1 mg/mL stock solution of the tested material, in ethanol, was added to each wells and mixed gently. The wells were closed and left at room temperature for 30 min. Finally, the absorbance was measured at 450 nm against a blank reagent by Beckman Coulter DTX 880 Multimode Detection System (Beckman Coulter, Brea, CA, USA). Ethanol was used as a negative control and BHT, BHA, and α-TOC were used as positive controls.

$$\text{Total antioxidant activity}(\%) = ((R_{\text{control}} - R_{\text{sample}}) / R_{\text{control}})) \times 100$$

2.6 Anticholinesterase activity

Acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) inhibitory activities were measured by a slightly modified spectrophotometric method, developed by Ellman et al. [15].

Acetylthiocholine iodide and butyryl thiocholine iodide were used as substrates of the reaction, and Ellman's Reagent, i.e. 5,5′-dithiobis-(2-ditrobenzoic acid) (DTNB) method, was applied for the measurement of the activity. Briefly, 130 μL of 100 mM sodium phosphate buffer (pH 8.0), 10 μL extract sample (4 mg/mL), and 20 μL of AChE or BChE solution were mixed and incubated for 15 min at 25 °C, after which 0.5 mM (15 μL) DTNB was added. The reaction was then initiated by addition of acetylthiocholine iodide (0.71 mM) or butyrylthiocholinechloride (0.2 mM). The hydrolysis of these extracts was monitored by UV-Vis spectrophotometer. Formation of yellow colored 5-thio-2-nitrobenzoate anion was followed at 412 nm as the result of the reaction of DTNB with thiocholine, released by an enzymatic hydrolysis of acetylthiocholine iodide or butyrylthiocholine chloride.

Ethanol was used as a solvent to dissolve extracts and the controls. Percentage inhibition of AChE and BChE was calculated using the following formula: $((E - S)/E \times 100)$. Ethanol was used as a blank control, and it is coded as E in the formula. Activity of the enzyme with the extract is coded as S. Galanthamine (4 mg/mL) was used as a positive control. All tests were conducted in triplicate.

2.7 Antibacterial and antifungal activity

Essential oils and methanol extracts of *S. metastasiantha* were tested against the following standard strains of *Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 25923, *Candida albicans* ATCC 10231, and *Mycobacterium smegmatis* for the determination of antimicrobial and antifungal activities.

The tube dilution procedure, outlined by the National Committee for Clinical Laboratory Standards, was used for the determination of antibacterial and antifungal activities of the extracts. All the tests were performed in Mueller-Hinton Broth. The methanol and chloroform extracts were evaporated and suspended in dimethyl sulfoxide (DMSO). The extracted essential oils were diluted with DMSO [16].

Final concentration of the essential oil was prepared from 4 to 2048 μg/mL in a well. Using McFarland turbidity standard, 0.5 McFarland bacteria suspension was inoculated per well in a 96-well round-bottom microplates. DMSO was used as a negative control for the activities. Oxacillin, gentamicin, streptomycin, and fluconazole were used as positive controls. All the tests were performed in triplicate, and the plates were incubated at 37 °C for 24–48 h, then, the endpoint was evaluated. The lowest concentration of the extracts, inhibiting the visible growth, was considered as the minimum inhibitory concentration [17].

3 Results and Discussion

Satureja metastasiantha was identified as a new species for the first time by Dr. Dirmenci and his group in the flora of Turkey. This species had previously been reported only in North of Iraq [7]. The form and smell of the species were found very similar to other commercially important *Satureja* species, which is rationally used as a spice in Turkey and Iraq. However, until now, there has been no chemical composition report available on that species in the literature. Therefore, herein, we aimed to determine the chemical composition of the essential oil of *S. metastasiantha* and its biological activity pattern.

Table 1: Essential oil of composition of *Satureja metastasiantha*.

RRI	Compounds	%	Identification method
855	2-Hexanal	0.1	RRI, MS
859	3-Hexen-1-ol	–	RRI, MS
927	Tricyclene	T	RRI, MS
930	α-Thujene	1.3	RRI, MS
939	α-Pinene	1.6	RRI, MS, ST
954	Camphene	1.0	RRI, MS
975	Sabinene	t	RRI, MS
979	β-Pinene	0.3	RRI, MS, ST
979	1-Octen-3-ol	0.2	RRI, MS
991	β-Myrcene	2.1	RRI, MS, ST
991	3-Octanol	0.1	RRI, MS
1003	α-Phellandrene	0.3	RRI, MS
1009	3-Carene	0.1	RRI, MS
1017	α-Terpinene	3.6	RRI, MS, ST
1025	***p*-Cymene**	**22.3**	**RRI, MS**
1029	Limonene	0.4	RRI, MS, ST
1031	1,8-Cineole	t	RRI, MS
1037	*trans*-β-Ocimene	0.2	RRI, MS
1050	*cis*-β-Ocimene	0.1	RRI, MS
1060	**γ-Terpinene**	**12.1**	**RRI, MS, ST**
1070	*cis*-Sabinene hydrate	0.4	RRI, MS
1096	Dimethylstyrene	0.2	RRI, MS
1097	Linalool	1.3	RRI, MS, ST
1141	*cis*-Verbenol	0.1	RRI, MS
1143	*cis*-Sabinol	t	RRI, MS
1145	*trans*-Verbenol	t	RRI, MS
1169	Borneol	2.2	RRI, MS, ST
1177	4-Terpineol	1.3	RRI, MS
1183	*p*-Cymen-8-ol	t	RRI, MS
1189	α-Terpineol	0.1	RRI, MS, ST
1196	Myrtenol	0.1	RRI, MS
1217	*trans*-Carveol	t	RRI, MS
1230	Nerol	t	RRI, MS
1238	β-Citral	t	RRI, MS, ST
1243	Carvone	0.7	RRI, MS
1253	Geraniol	t	RRI, MS, ST
1267	Geranial	t	RRI, MS
1291	**Thymol**	**21.0**	**RRI, MS**
1299	**Carvacrol**	**18.4**	**RRI, MS**
1359	Eugenol	–	RRI, MS
1362	Thymol acetate	0.1	RRI, MS
1373	Carvacrol acetate	–	RRI, MS
1377	α-Copaene	0.1	RRI, MS
1380	β-Bourbonene	0.1	RRI, MS
1391	β-Elemene	–	RRI, MS
1421	β-Ylangene	3.3	RRI, MS
1434	β-Gurjunene	0.1	RRI, MS
1441	Aromadendrene	0.6	RRI, MS
1480	γ-Muurolene	t	RRI, MS
1490	β-Selinene	0.1	RRI, MS
1496	Valencene	0.1	RRI, MS
1500	α-Muurolene	0.2	RRI, MS
1514	γ-Cadinene	–	RRI, MS
1523	δ-Cadinene	0.5	RRI, MS
1531	γ-Bisabolene	t	RRI, MS
1533	Nerolidol	0.1	RRI, MS
1539	α-Cadinene	0.3	RRI, MS
1546	α-Calacorene	t	RRI, MS
1578	Spathulenol	1.0	RRI, MS, ST
1583	Caryophyllene oxide	1.3	RRI, MS, ST
1654	α-Cadinol	t	RRI, MS, ST
	Total	99.5	

RRI, relative retention indices calculated against *n*-alkanes. % calculated from the FID chromatograms. T, trace (<0.1%). Identification methods: RI, identification based on the retention times (t_R) of genuine compounds on DB5 column; MS, identified on the basis of computer matching of the mass spectra with those of the Wiley and NIST and İLMER libraries and comparison with literature data. ST, co-injection with standard compounds. GC analyses were replicated three times (mean RSD% value is 0.05).

Sixty-one compounds were identified in the oil, distilled from the leaves of *S. metastasiantha*, representing 99.5% of the oil constituents. The main compounds found in the essential oil were characterized as *p*-cymene (22.3%), thymol (21.0%), carvacrol (18.4%), and γ-terpinene (12.1%). The other identified compounds and their percentages are given in Table 1.

When the chemical composition pattern of the species was compared with the other well studied and commercially important Anatolian *Satureja* species, *S. thymbra* and *S. cuneifolia*, an important difference was observed, which is the distribution ratio of carvacrol and thymol in the species. Thymol, carvacrol, and *p*-cymene compositions of the essential oil of the genus are important tools for chemotaxonomic evaluation of the genus. Thus, when we checked the previous reports of the species of the genus, we found that the thymol content was reported between 0.23% and 57.89%, while the carvacrol content was 2.39–62.82% [18] in *S. cuneifolia*, whereas the thymol and carvacrol percentages of *S. thymbra* was reported as 13.16% and 40.15%, respectively. The *p*-cymene content of those species were found in moderately low amounts, together with α-terpinene and γ-terpinene as expected, possibly due to the biosynthetic pathway of carvacrol and thymol through α-terpinene and γ-terpinene [18, 19].

In addition, the lower content of carvacrol, thymol, and high content of *p*-cymene were reported in *Satureja macrantha* and *S. intermedia* from Northern Iran [20, 21]. It was also observed in *S. hortensis* from Iran, which grows in strong continental climate regions [22]. The thymol and carvacrol contents of *S. hortensis* from different locations of Turkey were reported, and the geographical deviations

were discussed [23]. Moreover, changes in carvacrol content of *S. cuneifolia* during pre-flowering, flowering, and post-flowering periods were reported [24]. In that study, the thymol content of *S. hortensis* was reported as 0.1–43.0%. Finally, the chemical compositions of *S. spinosa* from Turkey, having one of the lowest level of thymol and carvacrol, were reported as 0.1–2.2% for carvacrol and 1.2% for thymol [25]. According to those data, we can clearly observe the chemotype variations of the species of Satureja in Turkey and Iran.

When looked at Table 1, as a result of relatively lower content of carvacrol and thymol, high content of *p*-cymene and γ-terpinene are observed. The lower amount of syntheses of carvacrol and thymol by species might be due to the strong continental climate conditions of the Hakkari province of Turkey similar to the Iranian species [22]. The environmental conditions and genetic patterns may affect the biosynthetic pathway of the species, and thus, higher contents of *p*-cymene and γ-terpinene were observed as chemotaxonomic markers of the species. *Satureja metastasiantha* could be considered as a new source of *p*-cymene and γ-terpinene together with carvacrol and thymol.

Apart from the first chemical report of the species, we also evaluated the antioxidant, acetylcholine, and butyrylcholine esterase activities of the essential oil and methanol extract of the species.

The antioxidant activities of the essential oils and the methanol extracts were determined by two methods, e.g. DPPH free radical scavenging activity and cupric ion reducing antioxidant capacity. DPPH free radical scavenging activities were determined by serial of concentrations of the tested materials in 10, 25, 50, and 100 μg/mL, where α-tocopherol, BHA, and BHT were used as positive controls. DPPH free radical scavenging activity results showed that the essential oil extracts do not have significant antioxidant capacity. The methanol extracts of 50 and 100 μg/mL displayed better antioxidant capacity than the other concentrations as expected because of possible higher content of phenolic compositions.

Antioxidant capacities of the essential oil and the extracts of the species in Trolox Equivalent Antioxidant Capacity values were determined by CUPRAC method via Cu^{2+}-neocuproine (Cu^{2+}-Nc) reagent as a chromogenic oxidizing reactant. The cupric ion (Cu^{2+}) reducing capability of the essential oil and methanol extract of the species are summarized in Table 2. Cupric ion reducing capacities of the essential oil, methanol extracts, and the standard compound, α-tocopherol, were determined. In spite of the radical scavenging capacity of the cupric ions, the reducing capacities of the essential oils and methanol extract were found moderately higher than DPPH, and the results were almost equal in both essential oil and methanol extract. However, the cupric ion (Cu^{2+}) reducing capacity of both was not found to be as powerful as α-tocopherol (Table 2).

AChE has an important role in the central nervous system, which catalyzes the cleavage of acetylcholine in the synaptic cleft after depolarization [26]. BChE is found in glial cells, neurons, and neuritic plaques [27]. Galanthamine is used frequently to treat the symptoms of Alzheimer's disease as an AChE inhibitor [28], which hydrolyses the acetylcholine compound involved in the communication between synapses in the nervous system. Thus, it is very important to cure Alzheimer's disease. BChE is synthesized in liver and released into the circulation. The fundamental role of BChE is the hydrolysis of succinylcholine, which is used as a local anesthetic and a short-term muscle relaxant before a general anesthesia in surgery. AChE has a rapid and effective hydrolysis capacity against a natural neurotransmitter, acetylcholine, while BChE can do this against synthetic substrates, mainly butyrylcholine [29].

If any problem arises in this pathway, then, Alzheimer's disease may take place, which could be treated using cholinesterase inhibitors to bring the acetylcholine level to normal. Thus, to find new AChE and BChE sources

Table 2: Antioxidant capacity of the essential oil and the methanol extract of *S. metastasiantha*.

Tested material	DPPH				$TEAC_{CUPRAC}$
	10 μg/mL	25 μg/mL	50 μg/mL	100 μg/mL	
Essential oil	0.5±0.6	1.6±0.5	3.4±1.2	10.7±0.6	0.52±0.09
MeOH extract	9.5±1.5	18.8±3.2	35.0±2.0	60.8±2.5	050±0.06
BHA	82.1±1.6	85.8±0.8	86.3±0.2	86.7±0.2	–
BHT	40.8±1.8	59.3±0.4	65.1±0.3	79.0±0.6	–
α-Tocopherol	63.0±0.02	85.9±0.02	86.2±0.02	86.1±0.02	0.95±0.06
DMSO	0.4±0.00	0.4±0.00	0.4±0.00	0.4±0.00	0.11±0.00

DPPH, 2,2-diphenyl-1-picrylhydrazyl; TEAC, Trolox equivalent antioxidant capacity; MeOH, methanol; BHA, butylated hydroxyanisole; BHT, butylated hydroxytoluene; DMSO, dimethyl sulfoxide.

Table 3: Anticholinesterase activity of the essential oil and the methanol extract of *S. metastasiantha*.

Tested materials	AChE % inhibition	BChE % inhibition
Essential oils	30 ± 2.1	18.1 ± 0.7
Methanol extract	29.9 ± 5.9	7.2 ± 2.8
Galantamine	96.8 ± 1.3	83.3 ± 0.7

Concentrations of all tested materials are used as 200 μg/mL. AChE, acetylcholinesterase; BChE, butyrylcholinesterase.

is very important, and one of the best sources is the plant Kingdom, especially unreported species such as *S. metastasiantha*. In this study, we found that the AChE inhibition levels of essential oil and methanol extract are almost the same but three times lower than the standard galantamine (Table 3). However, the essential oil content of the species was found more active than the methanol extract.

The antimicrobial activity of the essential oil and the methanol extract of the species were evaluated as MIC against *E. coli* ATCC 25922, *S. aureus* ATCC 25923, *C. albicans* ATCC 10231, and *M. smegmatis*. Unfortunately, only the essential oil of the species showed a moderate activity against *S. aureus* as 128 μg/mL in the tested organisms. The results of the antimicrobial and antifungal activity tests are given in Table S1 in the Supporting Information.

4 Conclusion

Herein, the chemical compositions of the essential oil of *S. metastasiantha* along with the antioxidant capacity, anticholinesterase (AChE), BChE, and antimicrobial activities and antifungal properties of the essential oil and methanol extract are reported for the first time. According to our results, the species could be considered as a new source of *p*-cymene, carvacrol, thymol, and γ-terpinene. Moderate antioxidant capacity and anticholinesterase and BChE activities were determined with essential oil and methanol extract of the species. However, any remarkable antimicrobial activity of the essential oil of the species, except *S. aureus*, was not observed (see Table S1). This lower antimicrobial activity of the essential oil of the species compared with the other members of the genus might be due to the result of the lower carvacrol and thymol contents.

References

1. Goren AC, Topcu G, Bilsel G, Bilsel M, Wilkinson JM, Cavanagh H. Analysis of essential oil of Satureja thymbra by hydrodistillation, thermal desorber, and headspace GC/MS techniques and its antimicrobial activity. Nat Prod Res 2004;18:189–95.
2. Goren AC. Use of Stachys species (Mountain Tea) as herbal tea and food. Rec Nat Prod 2014;2:71–82.
3. Celep F, Dirmenci T. Systematic and biogeographic overview of Lamiaceae in Turkey. Nat Volatiles Essent Oils 2017;4:14–27.
4. Harley RM, Atkins S, Budantsev A, Cantino PH, Conn B, Grayer R, et al. The families and genera of vascular plants, VII, flowering plants dicotyledons. In: Kubitzki K, editor. Berlin: Springer, 2004;7:167–275.
5. Goavaerts R, Paton A, Harvey Y, Navarro TP. World checklist of Lamiaceae, Kew. Facilitated by the Royal Botanic Gardens. http://apps.kew.org/wcsp/. Accessed: 29 October 2019.
6. Davis PH. Flora of Turkey and east Aegean Islands. Edinburgh: Edinburgh Univ. Press, 1982.
7. Dirmenci T, Yildiz B, Oztekin M. A new record for the flora of Turkey: Satureja metastasiantha Rech.f. (Lamiaceae). Bagbahce Bil Derg 2019;6:54–8.
8. Dirmenci T, Yazici T, Özcan T, Çelenk S, Martin E. A new species and a new natural hybrid of *Origanum* L. (Lamiaceae) from the west of Turkey. Turk J Bot 2018:73–90.
9. Rechinger KH, editors. Satureja L. Flora Iranica. Austria, 1980.
10. Baser KHC. Aromatic biodiversity among the flowering plant taxa of Turkey. Pure Appl Chem 2002;74:527–49.
11. Kurkcuoglu M, Tümen G, Baser KH. Essential oil constituents of Satureja boissieri from Turkey. Chem Nat Compd 2001;37:329–31.
12. Hatipoglu S, Zorlu N, Dirmenci T, Gören A, Ozturk T, Topcu G. Determination of volatile organic compounds in fourty five Salvia species by thermal desorption-GC-MS technique. Rec Nat Prod 2016;10:659–700.
13. Apak R, Güçlü K, Özyürek M, Esin Karademir S, Erçağ E. The cupric ion reducing antioxidant capacity and polyphenolic content of some herbal teas. Int J Food Sci Nutr 2006;57:292–304.
14. Han H, Yılmaz H, Gülçin I. Antioxidant activity of flaxseed (Linum usitatissimum L.) shell and analysis of its polyphenol contents by LC-MS/MS. Rec Nat Prod 2018;12:397–402.
15. Ellman GL, Courtney KD, Andres V, Featherstone RM. A new and rapid colorimetric determination of acetylcholinesterase activity. Biochem Pharmacol 1961;7:88–95.
16. Çıkrıkçı S, Mozioğlu E, Yılmaz H. Biological activity of curcuminoids isolated from Curcuma longa. Rec Nat Prod 2008;1:19–24.
17. National Committee for Clinical Laboratory Standards. Methods for dilution antimicrobial susceptibility test for bacteria that grow aerobically. Approved Standard M7-*A2*. Villanova: NCLSS, 1990.
18. Tumen G, Kirimer N, Ermin N, Baser KH. The essential oil of Satureja cuneifolia. Planta Med 1998;64:81–3.
19. Crocoll C. Biosynthesis of the phenolic monoterpenes, thymol and carvacrol, by terpene synthases and cytochrome P450s in oregano and thyme. PhD Thesis, Friedrich-Schiller-Universität, 2010.
20. Gohari AR, Hadjiakhoondi A, Sadat-ebrahimi E, Saeidnia S, Shafiee A. Composition of the volatile oils of Satureja spicigera C. Koch Boiss and S. macrantha C.A. Mey from Iran. Flavour Fragr J 2006;21:348–50.
21. Sefidkon F, Jamzad Z. Chemical composition of the essential oil of three Iranian Satureja species (S. mutica, S. macrantha and S. intermedia). Food Chem 2005;91:1–4.
22. Farzaneh M, Kiani H, Sharifi R, Reisi M, Hadian J. Chemical composition and antifungal effects of three species of Satureja (S. hortensis, S. spicigera, and S. khuzistanica) essential oils on the main pathogens of strawberry fruit. Postharvest Biol Technol 2015;109:145–51.

23. Baser KH, Ozek T, Plant M. A comparative study of the essential oils of wild and cultivated Satureja hortensis L. J Essent Oil Res 2004;16:422–4.
24. Koşar M, Demirci B, Demirci F, Başer KH. Effect of maturation on the composition and biological activity of the essential oil of a commercially important Satureja species from Turkey: Satureja cuneifolia Ten. (Lamiaceae). J Agric Food Chem 2008;56:2260–5.
25. Tabanca N, Kürkcüoglu M, Baser KH, Tümen G, Duman H. Composition of the essential oils of Satureja spinosa L. J. Essent Oil Res 2004;16:127–8.
26. Mohd W, Hakimi N, Salleh W, Ahmad F. Antioxidant and anticholinesterase activities of essential oil of Alseodaphne peduncularis Meisn. Turk J Pharm Sci 2016;13:347–50.
27. Giacobini E. Cholinesterases: new roles in brain function and in Alzheimer's disease. Neurochem Res 2003;28:515–22.
28. Khan MB, Palaka BK, Sapam TD, Subbarao N. Screening and analysis of acetyl-cholinesterase (AChE) inhibitors in the context of Alzheimer's disease. Bioinformation 2018;14:414–28.
29. Bayrak C, Taslimi P, Gulcin I, Menzek A. The first synthesis of 4-phenylbutenone derivative bromophenols including natural products and their inhibition profiles for carbonic anhydrase, acetylcholinesterase and butyrylcholinesterase enzymes. Bioorg chem 2017;72:359–66.

Supplementary Material: The online version of this article offers supplementary material (https://doi.org/10.1515/znc-2019-0208).