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## Sage-called plant species sold in Turkey and their antioxidant activities

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**Abstract:** Sage is commonly consumed as a herbal tea in Anatolia, where not only *Salvia* species, but also *Sideritis* species are called “sage” by the local people. Therefore, it was decided to investigate the most common species of sage-called plants sold in aktars (traditional herb-selling stores). Eighty-seven samples randomly purchased from 21 provinces throughout Turkey were identified, which finally led to the identification of 7 species; *Salvia tomentosa*, *Salvia fruticosa*, *Sideritis congesta*, *Sideritis pisidica* var. *termessi*, *Sideritis arguta*, *Sideritis perfoliata* and *Sideritis libanotica* subsp. *linearis*. Infusions prepared from all samples were preliminarily tested for their antioxidant activity and 7 representative species were further evaluated by the 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging, ferrous ion-chelating and ferric-reducing antioxidant power (FRAP) tests at 0.25, 0.50, and 1.0 mg ml<sup>-1</sup> and for their anti-acetylcholinesterase activity. The infusions were subjected to the DPPH bioautographic revelatory test, which led to the conclusion that a flavonoid derivative seemed to be responsible for the antioxidant activity in *S. congesta* and *S. pisidica* var. *termessi*.

**Keywords:** sage; *Salvia*; *Sideritis*; infusion; antioxidant; acetylcholinesterase.

### INTRODUCTION

In Turkey, sage is known as “adaçayı” and is commonly consumed as teas, which are also used in Anatolian folk medicine.<sup>1</sup> *Salvia* species (*Lamiaceae*) are known as “sage” in English, usually referring to *S. officinalis*, whereas throughout Turkey, species of several plant genera, *Salvia*, *Sideritis* and very rarely *Stachys*, are usually known as sage and sold in aktars (traditional herb-selling stores).<sup>2,3</sup> As these three genera belonging to *Lamiaceae* are similar to each other

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in terms of their morphology, they are easily confused by local people. In addition, as *Salvia* and *Sideritis* species are represented by approximately 90 and 40 species, respectively, in Turkey,<sup>4-6</sup> the aim of the current study was to survey which species (taxa) are sold as sage (adaçayı) in aktars and local bazaars. It was also decided to determine their antioxidant activity by three different *in vitro* methods, *i.e.*, by the 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging, ferrous ion-chelating, and ferric-reducing antioxidant power (FRAP) tests. Moreover, some *Salvia* species have also been reported for memory enhancement in European folk medicine.<sup>7</sup> Thus the inhibitory activity of the infusions towards acetylcholinesterase (AChE), the key enzyme in the pathogenesis of Alzheimer's disease (AD),<sup>8</sup> was also investigated using the spectrophotometric Ellman method.

#### EXPERIMENTAL

##### *Plant materials*

The name of the aktars and provinces, purchase date and identification of 87 sage-called plant materials which were sold as sage (adaçayı in Turkish) and used in this study are listed in Table I. The obtained materials (already in dried form) were identified by three taxonomists; namely Prof. Dr. Hayri Duman (Department of Biology, Faculty of Art and Science, Gazi University, Ankara), Dr. Gülderen Yılmaz (Department of Pharmaceutical Botany, Faculty of Pharmacy, Ankara University, Ankara, Turkey), and Dr. Ferhat Celep (Department of Biology, Middle East Technical University, Ankara, Turkey). The samples are kept at the Pharmacognosy Research Laboratory of the Faculty of Pharmacy, Gazi University, Ankara, Turkey.

TABLE I. Name of the aktars and provinces, purchase dates, and identification as well as the yields of the aqueous extract of the sage-called plant materials used in this study

Sample No.	Name of the aktars and province	Purchase date	Identification of the plant materials	Aqueous extract yields, % (w/w)
1	Yeniğül Vip Kuruyemiş, Ankara	2007	<i>Salvia tomentosa</i>	3.98
2	Tuzabat Köyü, Muğla	2007	<i>Salvia fruticosa</i>	3.93
3	Şekeroğlu Baharat, Kilis	2007	<i>Sideritis congesta</i>	9.76
4	Ulucami Baharatçısı, Bursa	2007	<i>Salvia fruticosa</i>	4.66
5	Etem Baharatları, Trabzon	2007	<i>Sideritis pispidica</i> var. <i>termessi</i>	15.0
6	Etem Baharatları, Trabzon	2007	<i>Salvia fruticosa</i>	11.4
7	Bağdat Ticaret Gıda Limited Şti., Ankara	2007	<i>Sideritis congesta</i>	8.26
8	Emre Baharat, Kastamonu	2007	<i>Sideritis congesta</i>	10.58
9	Gıda Pazarı, Ankara	2008	<i>Salvia fruticosa</i>	4.74
10	Yeni Belediye İş Merkezi, Çankiri	2007	<i>Salvia fruticosa</i>	6.03
11	Lokman Baharat, Karabük	2007	<i>Salvia fruticosa</i>	8.68
12	Ulukuş Alternatif Tıbbın Doğal Çaylar ve Şifalı Nubatat Bitkileri, Gaziantep	2007	<i>Sideritis congesta</i>	8.68
13	Yaşam Eren Baharat, Edirne	2007	<i>Salvia fruticosa</i>	4.14
14	Yayla Baharat, İstanbul	2008	<i>Salvia fruticosa</i>	6.51

TABLE I. Continued

Sample No.	Name of the aktars and province	Purchase date	Identification of the plant materials	Aqueous extract yields, % (w/w)
15	Özşen Lokman Hekim Baharat, Ankara	2008	<i>Sideritis congesta</i>	9.0
16	Arifoğlu Baharat, Çanakkale	2007	<i>Salvia fruticosa</i>	6.25
17	Sümbül Efendi Baharatçısı, İstanbul	2008	<i>Salvia fruticosa</i>	4.46
18	Aksel Baharat, İstanbul	2008	<i>Salvia fruticosa</i>	7.12
19	Özçiçek Kuruyemiş, İstanbul	2008	<i>Salvia fruticosa</i>	4.32
20	Şifa Sultan Bitkisel Ürünler, İstanbul	2008	<i>Salvia fruticosa</i>	7.54
21	İnan baharatları ve Aktar, İstanbul	2008	<i>Salvia fruticosa</i>	7.69
22	Mesut Güneş Aktar ve Baharat, İstanbul	2008	<i>Salvia fruticosa</i>	7.16
23	Muhittin Lokman Hekim, Ankara	2008	<i>Salvia fruticosa</i>	5.02
24	Muhittin Lokman Hekim, Ankara	2008	<i>Sideritis congesta</i>	12.02
25	Doğa Baharat Dünyası, İstanbul	2008	<i>Salvia fruticosa</i>	7.21
26	Nur Baharat, İstanbul	2008	<i>Salvia fruticosa</i>	4.89
27	Kırk Ambar, İstanbul	2008	<i>Salvia fruticosa</i>	8.57
28	Yeni Çavuşoğlu Baharatları, İstanbul	2008	<i>Salvia fruticosa</i>	2.04
29	Karabulut Şifa Baharatçısı, İstanbul	2008	<i>Salvia fruticosa</i>	5.09
30	Paşam Baharat (Mısır Çarşısı), İstanbul	2008	<i>Salvia fruticosa</i>	7.71
31	Gözde Baharat ve Kuruyemiş, İstanbul	2008	<i>Salvia fruticosa</i>	9.01
32	Lokman Baharat, İstanbul	2008	<i>Salvia fruticosa</i>	5.88
33	Atlar Baharat, İstanbul	2008	<i>Salvia fruticosa</i>	4.08
34	Hazerbaba, İstanbul	2008	<i>Sideritis congesta</i>	12.64
35	Antep Pazarı, İstanbul	2008	<i>Salvia fruticosa</i>	5.59
36	Uğur Kuruyemiş, İstanbul	2008	<i>Salvia fruticosa</i>	3.81
37	Sena Baharat, İstanbul	2008	<i>Salvia fruticosa</i>	5.10
38	Kardeşler Baharat, Kirikkale	2008	<i>Salvia fruticosa</i>	7.72
39	Ünlü Lokman Hekim, Kirikkale	2008	<i>Salvia fruticosa</i>	7.28
40	Coşkun Kuruyemiş, Ankara	2008	<i>Salvia fruticosa</i>	3.97
41	Hünkar Tohumculuk, Ankara	2008	<i>Salvia fruticosa</i>	6.58
42	Lokman Hekim, Ankara	2008	<i>Salvia fruticosa</i>	2.50
43	Avan Kuruyemiş ve Baharat, Ankara	2008	<i>Salvia fruticosa</i>	4.71
44	Paşa Süpermarket ve Mandıra, Ankara	2008	<i>Salvia fruticosa</i>	5.84
45	Safari Kuruyemiş, Ankara	2008	<i>Salvia fruticosa</i>	3.98
46	Akkaynak Süper Market, Ankara	2008	<i>Salvia fruticosa</i>	1.58
47	Coşkunlar Gıda, Ankara	2008	<i>Salvia fruticosa</i>	5.41
48	Zeyveli Gıda Pazarı, Ankara	2008	<i>Sideritis congesta</i>	6.49
49	Mert Gıda, Ankara	2008	<i>Sideritis congesta</i>	7.67

TABLE I. Continued

Sample No.	Name of the aktars and province	Purchase date	Identification of the plant materials	Aqueous extract yields, % (w/w)
50	Berat Baharatları, Ankara	2008	<i>Sideritis congesta</i>	9.90
51	Kuruyemiş Dünyası, Ankara	2008	<i>Sideritis congesta</i>	15.0
52	İmalatçı Erhan Zeytincilik, Ankara	2008	<i>Salvia fruticosa</i>	5.01
53	Erpaş Gıda, Ankara	2008	<i>Salvia fruticosa</i>	6.57
54	Şifa Baharatları, Elazığ	2008	<i>Salvia fruticosa</i>	4.65
55	Özgıda, Elazığ	2008	<i>Sideritis congesta</i>	12.60
56	Efka Baharat, Muğla	2008	<i>Salvia fruticosa</i>	6.19
57	Seçkin Manav, Muğla	2008	<i>Salvia fruticosa</i>	5.36
58	Balcı Gökmen, Muğla	2008	<i>Salvia fruticosa</i>	4.30
59	Merve Gıda, Çorum	2008	<i>Salvia fruticosa</i>	5.53
60	Ebru Kuruyemiş, Bingöl	2008	<i>Salvia fruticosa</i>	7.13
61	Nur Gıda Pazarı, Bingöl	2008	<i>Salvia fruticosa</i>	3.30
62	Hatemoğlu Kuruyemiş, Ankara	2008	<i>Sideritis congesta</i>	12.50
63	Arıvital, Ankara	2008	<i>Salvia fruticosa</i>	4.53
64	Gizem Lokman Hekim, Ankara	2008	<i>Salvia fruticosa</i>	3.06
65	Can Baharat, Ankara	2008	<i>Salvia fruticosa</i>	5.83
66	Çağrı Baharat, Ankara	2008	<i>Sideritis congesta</i>	12.41
67	Ünlü Lokman, Ankara	2008	<i>Sideritis congesta</i>	10.0
68	Ünlü Lokman, Ankara	2008	<i>Salvia fruticosa</i>	9.36
69	Sabancı Kemal Hoca, Ankara	2008	Could not be identified	9.97
70	Dr. Ali Nazmi Lokman Hekim, Ankara	2008	<i>Sideritis congesta</i>	8.65
71	Candan Lokman, Adana	2008	<i>Salvia fruticosa</i>	8.72
72	Esen Lokman Baharat, Adana	2008	<i>Salvia fruticosa</i>	3.05
73	Çerçi Hüsnü Yusuf, Adana	2008	<i>Salvia fruticosa</i>	8.84
74	Ağar Bakkaliyesi, Adana	2008	<i>Salvia fruticosa</i>	5.76
75	Kantarmacılar Çerçi Mehmet, Adana	2008	<i>Salvia fruticosa</i>	7.71
76	Kantarmacılar Çerçi Mehmet, Adana	2008	<i>Sideritis congesta</i>	9.93
77	Has Çerçi Yusuf, Adana	2008	<i>Salvia fruticosa</i>	8.52
78	Çerçi Uğur Yusuf Ticaret, Adana	2008	<i>Salvia fruticosa</i>	2.42
79	Metin Baharat, Sivas	2008	<i>Sideritis congesta</i>	8.58
80	Özfidan Baharat, Sivas	2008	<i>Sideritis congesta</i>	8.42
81	Mevsim Ticaret, Sivas	2008	<i>Sideritis congesta</i>	12.26
82	Köylüoğlu Baharat, Afyon	2008	<i>Salvia fruticosa</i>	3.58
83	Karaca Baharat, Afyon	2008	<i>Salvia fruticosa</i>	6.86
84	Kamburoğlu Baharat, Afyon	2008	<i>Salvia fruticosa</i>	2.33
85	Oktaş Baharat, Antalya	2008	<i>Sideritis perfoliata</i>	18.50
86	Halk Pazarı, Antalya	2008	<i>Sideritis arguta</i>	5.53
87	Mustafa Şimşek Baharatçı, Antalya	2008	<i>Sideritis libanotica</i> subsp. <i>linearis</i>	2.62

### Preparation of the infusions

Approximately 10 g from each of the 87 samples sold as “sage” was weighed accurately on a digital balance and 150 ml of boiling distilled water was poured onto each sample and left for 10 min at room temperature in order to prepare the infusions, which is in accordance with the traditional method for sage tea preparation in Anatolia. The aqueous parts of each infusion was filtrated and lyophilized. The lyophilized extracts, the yields (w/w) of which are given in Table I, were employed in the AChE inhibitory and antioxidant activity tests.

### Antioxidant activity tests

**DPPH radical scavenging activity.** The stable DPPH radical scavenging activity was determined by the Blois method.<sup>9</sup> Gallic acid and butylated hydroxyanisol (BHA) were employed as the references. Inhibition of DPPH (*I*) in percent was calculated as:

$$I (\%) = 100((A_{\text{blank}} - A_{\text{sample}}) / A_{\text{blank}}) \quad (1)$$

where  $A_{\text{blank}}$  is the absorbance of the control reaction (containing all reagents except the test sample) and  $A_{\text{sample}}$  is the absorbance in the presence of the extracts/reference. The analyses were run in triplicate and the results are expressed as average values with the standard error mean (SEM).

**Ferrous ion-chelating effect.** The ferrous ion-chelating effect of all the infusions was estimated by the method of Chua.<sup>10</sup> The ratio of inhibition of the formation of the ferrozine- $\text{Fe}^{2+}$  complex was calculated by Eq. (1), where  $A_{\text{blank}}$  is the absorbance of the control reaction (containing only  $\text{FeCl}_2$  and ferrozine), and  $A_{\text{sample}}$  is the absorbance in the presence of the extracts/reference. The analyses were run in triplicate and the results are expressed as average values with the standard error mean (SEM).

**Ferric-reducing antioxidant power assay (FRAP).** The ferric-reducing power (FRAP) of the infusions was tested using the assay of Oyaizu.<sup>11</sup> The analyses were performed in triplicate. Increased absorbance of the reaction mixture indicated increased reducing power.

### Bioautographic DPPH revelatory test

The infusions belonging to *Salvia tomentosa*, *Salvia fruticosa*, *Sideritis congesta*, *Sideritis pisidica* var. *termessi*, *Sideritis arguta*, *Sideritis perfoliata* and *Sideritis libanotica* subsp. *linearis* were subjected to thin layer chromatography (TLC) using a solvent system consisting of ethyl acetate:methanol:water (60:20:5). The plate after drying at room temperature was examined under UV 254 and 366 nm light and sprayed with 4 % DPPH solution. The radical scavenging spots turned to a yellow color on a purple background.

### Analysis of the active spots in the bioautographic DPPH revelatory test

The spots which were found to be active in the DPPH revelatory test were subjected to TLC again using the same solvent system and treated with three different spraying agents: namely 5 % sulfuric acid, 1 % vanillin and naturstoff reagent in order to obtain preliminary information about phytochemical nature of the compounds.

### Anti-AChE activity

Anti-AChE activity was assayed by the spectrophotometric method of Ellman.<sup>12</sup> Electric eel AChE (Type-VI-S, EC 3.1.1.7, Sigma) was used as the enzyme source, while acetylthiocholine iodide (Sigma, St. Louis, MO, USA) was employed as the substrate of the reaction. 5,5'-Dithio-bis(2-nitrobenzoic)acid (DTNB, Sigma, St. Louis, MO, USA) was used for the measurement of the cholinesterase activity. All the other reagents and conditions were same as

described in a previous publication.<sup>13</sup> The experiments were performed in triplicate. Galanthamine (Sigma, St. Louis, MO, USA) was the reference.

#### *Total phenol and total flavonoid contents*

Phenolic contents of the extracts were assayed according to the Folin-Ciocalteu method.<sup>14</sup> The samples were mixed with Folin-Ciocalteu reagent (Sigma) and sodium carbonate (7.5 %). After incubation at 40 °C for 30 min, the absorption was measured at 760 nm. The total phenolic contents are expressed as gallic acid equivalents (GAE, mg g<sup>-1</sup> extract). The total flavonoid contents were calculated by the aluminum chloride colorimetric method.<sup>15</sup> The absorbance of the reaction mixture was measured at 415 nm.

### RESULTS AND DISCUSSION

In this study, the species of 87 plant samples, which were sold as “sage”, were identified as *Salvia fruticosa* (61 samples), *Sideritis congesta* (20 species), *Salvia tomentosa* (1 species), *Sideritis pisidica* var. *termessi* (1 species), *Sideritis arguta* (1 species), *Sideritis perfoliata* (1 species), *Sideritis libanotica* subsp. *linearis* (1 species) and one unidentified species. In accordance with the traditional method of preparing sage tea in Anatolia, infusions from the 87 samples were prepared and the preliminary antioxidant activity of all infusions was determined using the DPPH radical scavenging test at a concentration of 1.0 mg ml<sup>-1</sup>. Their radical scavenging activity against DPPH was found to change between 12.69 and 71.39 % (Table II). Further antioxidant activity tests were only performed on the infusions of the 7 identified species given above using DPPH radical scavenging, ferrous ion-chelating and ferric-reducing antioxidant power (FRAP) tests at concentrations of 0.25, 0.50, and 1.0 mg ml<sup>-1</sup>. Among them, the *S. fruticosa* (65.04 %), *S. arguta* (60.70 %), and *S. congesta* (59.90 %) infusions had the highest scavenging effect towards DPPH (Table III), while *S. fruticosa* (2.303) and *S. arguta* (1.190) gave the best FRAP results (Table IV). All infusions displayed an insignificant effect in the ferrous ion-chelating tests (Table IV). These 7 infusions were tested for their *in vitro* acetylcholinesterase inhibition at concentrations of 0.25, 0.50, and 1.0 mg ml<sup>-1</sup> using an ELISA microplate reader and were found to exert no activity in this assay.

The calibration equations for the total phenol and flavonoid contents of the infusions were found to be  $y = 0.0009x + 0.0018$  ( $r^2 = 0.87$ ) and  $y = 2.0447x - 0.0302$  ( $r^2 = 0.99$ ), respectively. Accordingly, the richest infusion in terms of both total phenol and total flavonoid contents belonged to *S. congesta* (Table III). Subsequently, the 7 infusions were monitored by TLC and post spraying with a 4 % DPPH solution. Only two spots having DPPH radical scavenger activity, evidenced by a yellow color on a purple background, were found, belonging to the infusions of *S. congesta* and *S. pisidica* var. *termessi*. Those spots possessed the same  $R_f$  value, which seemed to be the same compound, and were sprayed with 5 % sulfuric acid, 1 % vanillin and naturstoff reagents. The spots became bright yellow with 1 % sulfuric acid and the naturstoff reagent, which led to the con-

clusion that a flavonoid derivative was responsible for the antioxidant activity of the infusions of *S. congesta* and *S. pisidica* var. *termessi*.

TABLE II. DPPH free radical scavenging activity as percentage of inhibition $\pm$ SEM (standard error mean), %, of the aqueous extracts of the sage-called species at 1.0 mg/ml

Sample No.	Value	Sample No.	Value	Sample No.	Value
1	34.01 $\pm$ 1.01	32	71.12 $\pm$ 0.18	63	37.11 $\pm$ 0.85
2	48.51 $\pm$ 1.79	33	32.49 $\pm$ 1.17	64	55.07 $\pm$ 0.32
3	47.23 $\pm$ 0.44	34	46.80 $\pm$ 0.21	65	50.45 $\pm$ 0.52
4	52.31 $\pm$ 1.47	35	41.73 $\pm$ 0.10	66	48.74 $\pm$ 1.69
5	36.76 $\pm$ 1.84	36	21.12 $\pm$ 1.00	67	35.81 $\pm$ 0.11
6	63.14 $\pm$ 1.72	37	46.46 $\pm$ 0.64	68	45.31 $\pm$ 1.05
7	39.35 $\pm$ 1.72	38	53.59 $\pm$ 1.08	69	88.23 $\pm$ 1.30
8	56.86 $\pm$ 0.32	39	27.84 $\pm$ 0.49	70	61.33 $\pm$ 1.16
9	43.00 $\pm$ 0.57	40	39.59 $\pm$ 1.39	71	45.16 $\pm$ 1.05
10	32.69 $\pm$ 1.07	41	28.69 $\pm$ 0.11	72	65.04 $\pm$ 0.65
11	33.46 $\pm$ 0.74	42	58.02 $\pm$ 0.43	73	50.75 $\pm$ 0.74
12	66.40 $\pm$ 0.32	43	45.01 $\pm$ 0.42	74	51.12 $\pm$ 1.05
13	57.83 $\pm$ 0.63	44	47.40 $\pm$ 0.63	75	51.42 $\pm$ 0.21
14	40.41 $\pm$ 1.06	45	42.03 $\pm$ 0.21	76	41.14 $\pm$ 1.27
15	56.57 $\pm$ 0.32	46	42.40 $\pm$ 0.52	77	46.28 $\pm$ 1.15
16	63.71 $\pm$ 0.74	47	46.95 $\pm$ 0.84	78	12.69 $\pm$ 0.32
17	17.04 $\pm$ 1.02	48	50.84 $\pm$ 1.07	79	34.88 $\pm$ 0.63
18	34.48 $\pm$ 0.63	49	59.90 $\pm$ 1.67	80	36.04 $\pm$ 0.11
19	22.14 $\pm$ 1.55	50	64.16 $\pm$ 1.37	81	32.49 $\pm$ 0.21
20	52.71 $\pm$ 0.90	51	55.44 $\pm$ 0.21	82	34.43 $\pm$ 0.15
21	70.12 $\pm$ 1.12	52	53.72 $\pm$ 0.73	83	59.66 $\pm$ 1.37
22	64.78 $\pm$ 1.49	53	29.36 $\pm$ 0.70	84	23.25 $\pm$ 1.47
23	38.54 $\pm$ 0.62	54	23.60 $\pm$ 1.28	85	43.39 $\pm$ 1.68
24	61.67 $\pm$ 1.89	55	55.89 $\pm$ 1.05	86	60.70 $\pm$ 0.11
25	71.39 $\pm$ 0.10	56	29.05 $\pm$ 0.31	87	16.27 $\pm$ 0.10
26	42.62 $\pm$ 0.93	57	62.67 $\pm$ 0.54		
27	47.79 $\pm$ 1.94	58	40.76 $\pm$ 0.95		
28	16.19 $\pm$ 1.04	59	48.36 $\pm$ 0.31		
29	42.70 $\pm$ 0.32	60	31.15 $\pm$ 1.64		
30	47.77 $\pm$ 0.53	61	38.75 $\pm$ 0.63		
31	28.22 $\pm$ 0.99	62	50.45 $\pm$ 0.53		
References					
Gallic acid				92.57 $\pm$ 0.10	
Butylated hydroxyanisol (BHA)				81.60 $\pm$ 1.67	

The antioxidant and anti-AChE activities of the infusions obtained from the 7 representative plant species were also determined in order to find out if there were any differences between them. The infusions of the seven taxa identified as *S. fruticosa*, *S. congesta*, *S. tomentosa*, *S. pisidica* var. *termessi*, *S. arguta*, *S. perfoliata*, and *S. libanotica* subsp. *linearis* showed no inhibitory effect against

AChE. Previous research revealed that the components of sage which are active against AChE were the monoterpenes 1,8-cineole and  $\alpha$ -pinene found in major amounts in the essential oils.<sup>16</sup> The ineffectiveness of the infusions towards AChE could be that the essential oil and its components are not soluble in water.

TABLE III. Total phenol and total flavonoid contents, and DPPH free radical scavenging activity (inhibition percentage $\pm$ SEM (standard error mean)) of the aqueous extracts of 7 identified taxa out of 87 samples of sage-called species

Specimen	Total phenol content <sup>a</sup> $\pm$ SEM <sup>b</sup> , %	Total flavonoid content <sup>c</sup> $\pm$ SEM, %	Percentage of inhibition $\pm$ SEM against DPPH radical, %		
			DPPH concentration, mg ml <sup>-1</sup>		
			0.25	0.50	1.0
<i>Salvia tomentosa</i>	87.87 $\pm$ 0.32	46.31 $\pm$ 2.35	9.53 $\pm$ 0.11	33.28 $\pm$ 1.20	34.01 $\pm$ 1.01
<i>Salvia fruticosa</i>	129.94 $\pm$ 0.62	71.66 $\pm$ 3.14	22.27 $\pm$ 0.74	47.23 $\pm$ 1.33	65.04 $\pm$ 0.65
<i>Sideritis arguta</i>	88.09 $\pm$ 2.52	63.23 $\pm$ 1.11	14.09 $\pm$ 0.10	25.72 $\pm$ 1.38	60.70 $\pm$ 0.11
<i>Sideritis congesta</i>	154.10 $\pm$ 2.60	138.75 $\pm$ 2.94	17.54 $\pm$ 0.10	35.16 $\pm$ 0.11	59.90 $\pm$ 1.67
<i>Sideritis libanotica</i>	55.64 $\pm$ 1.26	35.94 $\pm$ 1.25	8.33 $\pm$ 0.11	10.27 $\pm$ 0.31	16.27 $\pm$ 0.10
<i>Sideritis</i> subsp. <i>linearis</i>					
<i>Sideritis perfoliata</i>	88.74 $\pm$ 2.44	42.49 $\pm$ 0.52	16.19 $\pm$ 0.85	33.28 $\pm$ 1.20	43.39 $\pm$ 1.68
<i>Sideritis pisi-dica</i> var. <i>termessi</i>	119.72 $\pm$ 1.34	96.56 $\pm$ 2.24	9.75 $\pm$ 0.21	21.52 $\pm$ 0.96	36.76 $\pm$ 1.84
References					
Gallic acid			ND <sup>d</sup>	91.61 $\pm$ 0.06	92.57 $\pm$ 0.10
BHA			ND	77.99 $\pm$ 0.48	81.60 $\pm$ 1.67

<sup>a</sup>Data expressed in mg equivalent of gallic acid (GAE) to 1 g of extract; <sup>b</sup>standard error mean; <sup>c</sup>data expressed in mg equivalent of quercetin to 1 g of extract; <sup>d</sup>not determined

On the other hand, the preliminary antioxidant activity screening of the infusions at a concentration of 1.0 mg ml<sup>-1</sup> by the DPPH radical scavenging test displayed a great variance with percentage if inhibition values from 12.69 to 71.39 %. These results clearly indicate that the sage-called plant samples had different phytochemical contents. Since the plant samples were sold in aktars under no serious official authority inspection, the collection time, date and name of the identifier are not clear. In addition, the preserving and storage conditions were not good, hence, some plant samples may have been exposed to direct sunlight or kept in open sacks. Thus, all these factors could affect the phytochemical content of the plants. Consistently, the analysis of the total phenol and flavonoid contents of the sage-called samples also showed remarkable variations (Table III).

In the further antioxidant assays for the 7 representative plant species, the *S. fruticosa* infusion exerted the best activity in the DPPH radical scavenging and FRAP assays (Tables III and IV). Several studies also showed significant antioxidant activity of the polar (water, ethanol, methanol, etc.) extracts of *S. fruticosa*, which was attributed to the existence of phenolic compounds, including rosmarinic acid, caffeic acid, carnosol, apigenin, and luteolin.<sup>17–20</sup> Accordingly, it could be speculated that the high antioxidant activity of this species could result from the presence of similar phenolic compounds in the infusion. However, not all the *S. fruticosa* samples purchased for this study displayed high radical scavenging activity (Table I). This variation could again depend on when and where the samples were obtained by the aktars as well as the duration of the storage time. Interestingly, in the DPPH bioautographic test, 7 infusions were subjected to TLC under the same conditions but only two infusions (*S. congesta* and *S. pisdica* var. *termessi*) were found to contain two active components, the colors of which turned into yellow on a purple background. In the TLC assay, chlorogenic acid, caffeic acid, gallic acid, rutin, quercetin, and kaempferol were used as references. However the two spots had different  $R_f$  value to those of the references. Therefore, the spots were sprayed separately with 5 % sulfuric acid, 1% vanillin, and naturstoff reagents. The color of flavonoid derivatives is known to become bright yellow with sulfuric acid and naturstoff reagents, which were in accordance with the present results. The vanillin reagent is more specific in revealing terpenic substances. Consequently, this led to the consideration that the active component of the above-mentioned *Sideritis* infusions could be a flavonoid derivative.

TABLE IV. Ferric ion-chelating effect and ferric-reducing antioxidant power (FRAP, absorbance at 700 nm±SEM) of the aqueous extracts of 7 identified taxa out of 87 samples of sage-called species

Aqueous extracts	Ferric ion-chelating capacity (inhibition±SEM <sup>a</sup> , %)			Ferric-reducing antioxidant power (absorbance at 700 nm±SEM)		
	Ferric ion concentration, mg ml <sup>-1</sup>					
	0.25	0.50	1.0	0.25	0.50	1.0
<i>Salvia tomentosa</i>	– <sup>b</sup>	2.59±0.45	19.35±0.40	0.38±0.01	0.68±0.08	1.12±0.01
<i>Salvia fruticosa</i>	–	3.70±0.97	6.95±1.70	0.96±0.02	1.64±0.05	2.30±0.05
<i>Sideritis arguta</i>	–	7.78±0.78	12.04±1.24	0.49±0.06	0.66±0.12	1.19±0.09
<i>Sideritis congesta</i>	–	2.50±0.65	5.56±0.88	0.50±0.01	0.67±0.06	1.02±0.08
<i>Sideritis libanotica</i> subsp. <i>linearis</i>	4.82±0.83	8.15±0.66	12.87±0.65	0.26±0.01	0.44±0.04	0.58±0.13
<i>Sideritis perfoliata</i>	–	13.52±0.52	20.84±3.27	0.42±0.02	0.86±0.03	1.03±0.09
<i>Sideritis pisdica</i> var. <i>termessi</i>	–	–	6.30±1.01	0.33±0.01	0.70±0.06	0.78±0.13
		Reference				
BHA	ND <sup>c</sup>	21.71±1.10	26.94±1.48	2.49±0.01	ND	ND

<sup>a</sup>Standard error mean; <sup>b</sup>no activity; <sup>c</sup>not determined

## CONCLUSIONS

The current survey showed that 7 plant species (*Salvia fruticosa*, *Sideritis congesta*, *Salvia tomentosa*, *Sideritis pisidica* var. *termessi*, *Sideritis arguta*, *Sideritis perfoliata* and *Sideritis libanotica* subsp. *linearis*) are sold under the name “sage” in aktars (traditional herbal stores) in Turkey. In previous reports, some *Stachys* species were also recorded to be known as sage in some parts of Turkey by local people. Since sage has been used for simple disorders in the Anatolian folk medicine, correct identification of the plant species is quite important in terms of their biological activity and phytochemical content. The present data showed that the most common species known as sage in Turkey are *S. fruticosa* and *S. congesta* and their antioxidant activity displays a great variation depending on diverse factors. Thus, the obtained results underline the importance and necessity of a serious inspection on plant species sold in aktars by authorities such as the Ministry of Health from the viewpoint of human health.

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## ИЗВОД

ВРСТЕ ЖАЛФИЈЕ ПОРЕКЛОМ ИЗ ТУРСКЕ И ЊИХОВА  
АНТИОКСИДАТИВНА АКТИВНОСТILKAY ERDOGAN-ORHAN<sup>1</sup>, ELIF BAKI<sup>1,2</sup>, SEZER ŞENOL<sup>1</sup> и GÜLDEREN YILMAZ<sup>3</sup>

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Жалфија се уобичајено користи за чај у Анадолији и то не само врсте *Salvia*, већ и врсте *Sideritis*. Проучавали смо биљке које се сматрају жалфијом, а продају се у локалним продавницама. Осамдесет седам узорака насумично купљене жалфије у 21 провинцији Турске је идентификовано и систематизовано у 7 врста: *Salvia tomentosa*, *Salvia fruticosa*, *Sideritis congesta*, *Sideritis pisidica* var. *termessi*, *Sideritis arguta*, *Sideritis perfoliata* и *Sideritis libanotica* subsp. *linearis*. Екстракти свих узорака су прелиминарно тестирани ради утврђивања антиоксидативне активности, а 7 типичних узорака је даље анализирано тестовима: а) DPPH, за одређивање слободних радикала, б) FRAP, за одређивање способности хелатирања феро-јона и редукције фери-јона и в) тестом за одређивање анти-ацетилхолинестеразне активности. Екстракти су подвргнути DPPH биоаутографском тесту, чији су резултати довели до закључка да су флавоноидни деривати одговорни за антиоксидативну активност врста *S. congesta* и *S. pisidica* var. *termessi*.

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## REFERENCES

1. T. Baytop, *Therapy with Medicinal Plants in Turkey (past and present)*, Istanbul University Publications No. 3255, Nobel Presshouse, Istanbul, 1999, p. 211

2. E. Sezik, N. Ezer, *Doğa Bilimleri Dergisi* **7** (1983) 163 (in Turkish)
3. G. Yilmaz, A. Güvenç, *J. Fac. Pharm. Ankara Univ.* **36** (2007) 87
4. I. C. Hedge, in *Flora of Turkey and the East Aegean Islands*, Vol. 10, P. H. Davis, Ed., Edinburgh University Press, Edinburgh, 1988, p. 400
5. A. Huber-Morath, in *Flora of Turkey and the East Aegean Islands*, Vol. 10, P. H. Davis, Ed., Edinburgh University Press, Edinburgh, 1988, p. 178
6. H. Duman, in *Flora of Turkey and the East Aegean Islands*, Vol. 11, A. Güner, N. Özhatay, T. Ekim, K. H. C. Başer, Eds., Edinburgh University Press, Edinburgh, 2000
7. N. Perry, G. Court, N. Bidet, J. Court, E. K. Perry, *Int. J. Ger. Psych.* **11** (1996) 1063
8. L. S. Schneider, *Clin. Geriatr. Med.* **17** (2001) 337
9. M. S. Blois, *Nature* **181** (1958) 1199
10. M. T. Chua, Y. T. Tung, S. T. Chang, *Biores. Technol.* **99** (2008) 1918
11. M. Oyaizu, *Jpn. J. Nutr.* **44** (1986) 307
12. G. L. Ellman, K. D. Courtney, V. Andres, R. M. Featherstone, *Biochem. Pharmacol.* **7** (1961) 88
13. F. S. Şenol, I. Orhan, F. Celep, A. Kahraman, M. Dogan, G. Yilmaz, B. Şener, *Food Chem.* **120** (2010) 34
14. V. L. Singleton, J. A. Rossi Jr., *Am. J. Enol. Viticult.* **16** (1965) 144
15. R. Woisky, A. Salatino, *J. Apicol. Res.* **37** (1998) 99
16. S. U. Savelev, E. J. Okello, E. K. Perry, *Phytother. Res.* **18** (2004) 315
17. A. Yildirim, A. Mavi, M. Oktay, A. A. Kara, Ö. F. Algur, V. Bilaloğlu, *J. Agric. Food Chem.* **48** (2000) 5030
18. K. Triantaphyllou, G. Blekas, D. Boskou, *Int. J. Food. Sci. Nutr.* **52** (2001) 313
19. V. Exarchou, N. Nenadis, M. Tsimidou, I. P. Gerothanassis, A. Troganis, D. Boskou, *J. Agric. Food Chem.* **50** (2002) 5294
20. L. Pizzale, R. Bortolomeazzi, S. Vichi, E. Überegger, L. S. Conte, *J. Sci. Food Agric.* **82** (2002) 1645.