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ANTIOXIDANT AND FREE RADICAL SCAVENGING ACTIVITIES OF EIGHT *Salvia* SPECIES*

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Lipid peroxidation is one of the major factors that cause deterioration during the storage and processing of foods. Synthetic antioxidants, such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and propylgallate (PG) are widely used; however, their use in food products is being questioned. Consumers have also became more cautious about the nutrition quality and safety of food additives. Antioxidants of natural origin, therefore, have drawn more attention [1]. Many spices and herbs are found to be potent sources of natural antioxidants. Among the spices reported to have antioxidant activity, rosemary and sage are well known [2, 3].

Salvia (*Lamiaceae*) represents one of the most diverse genera of plants in Turkey with 88 species of which 51% are endemic [4]. *Salvia* species have long been used as herbal tea and in a variety of food preparations. Preparations from *Salvia officinalis* (sage) are used for their medicinal properties, such as antispasmodic and antiseptic [5, 6]. *Salvia* species mainly contain essential oil and phenolic compounds such as flavonoids, phenolic acids, and phenolic diterpenes [7-9]. *Salvia* species have received particular attention as a source of natural antioxidants [8-13]. The main antioxidant activity of *S. officinalis* was reported to be attributed mainly to its phenolic compounds, such as carnosic acid, carnasol, and rosmarinic acid [12, 13].
The objective of this work was to evaluate the antioxidant activity of methanol extracts from eight *Salvia* species in different *in vitro* antioxidant test systems.

Aerial parts of the plant materials were collected from different regions of Turkey. Voucher specimens are kept at the Herbarium of the Faculty of Pharmacy (ESSE), Anadolu University, Eskisehir. Information on collection sites are given in Table 1. Butylated hydroxytoluene (BHT) and linoleic acid were purchased from Sigma Chemical Co. 1,1-Diphenyl-1-picrylhydrazyl radical (DPPH) was obtained from Aldrich Chemical Co. All the solvents used for extraction and antioxidant activity studies were of analytical grade. Crude sunflower oil was kindly provided by Demircanlar CO., Eskisehir, Turkey.
Powdered plant samples were continuously extracted in a Soxhlet extractor with methanol for 8 hours. Methanol was evaporated to dryness under vacuum at 40°C. Extraction yields are presented in Table 1.

Antioxidant Activity in Fe^{+2}-Induced Linoleic Acid System. Antioxidant activities of methanolic extracts of *Salvia* species at concentrations of 0.02% and 1% linoleic acid using the method of Fe^{+2}-induced linoleic acid peroxidation-TBA reactive substances were determined [14]. None of the extracts showed activity at a level of 0.02%. Although all the extracts exhibited antioxidant activity at a concentration of 1%, their inhibition ratios [Inhibition (%) = $(A_{control}-A_{sample}) / A_{control} \times 100$] were lower than that of BHT. Among them (Table 2) the antioxidant activity was in the order BHT > *S. chrysophylla* > *S. cilicica* ~ *S. tomentosa* > *S. halophila* > *S. fruticosa* ~ *S. crypthantha* > *S. sclarea* > *S. palaestina*.

Antioxidant Activity Testing by the Rancimat Method. Antioxidant activities of the methanolic extracts of *Salvia* species were also determined by the Rancimat Method (A 743 Rancimat apparatus, Metrohm AG, Switzerland) at concentrations of 0.02% and 1% (Table 2). A higher induction index indicates higher antioxidant activity.

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TABLE 1. Botanical Names and Collection Sites of Salvia Species Studied

Species	Collection site (date)	ESSE	Yield (%)
S. halophila Hedge*	Konya; Karabuk-Eskil 1 km, 890 m (July 1995)	1553	19.0
S. tomentosa Miller	Eskisehir; Bozdag (July 1997)	12381	17.1
S. fruticosa Miller	Antalya; Kas-Fethiye, 80 m (June 1995)	11361	26.3
S. chrysophylla Stapf*	Antalya; Elmali, Tekke village, Ciglikara, 1500 m (June	11312	21.7
S. sclarea L.	1995)	8935	20.1
S. clicica Boiss. & Kotscky*	Eskisehir; Bozdag, Ilica-Saricakaya (July 1997)	11552	12.8
S. crypthantha Montbret & Aucher ex Bentham*	Nigde; Ulukisla-Pozanti, 22 km, 1000 m (July 1995)	8939	18.3
S. palaestina Bentham	Eskisehir; Kanlipinar (May 1990)	13330	17.1
	Malatya; Yazihankaraca village (June 2000)		

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Endemic species.

TABLE 2. Antioxidant Activity of Salvia Extracts as Measured by the TBA and Rancimat Method

Sample	TBA Method Inhibition (%)[1,3]	Rancimat Method Induction Index[2,3]	
		0.02%	1%
S. chrysophylla	61.3±3.3	1.08±0.08	1.14±0.04
S. tomentosa	58.6±5.3	1.03±0.02	1.15±0.05
S. clicica	58.6±3.4	not active	not active
S. halophila	56.6±5.1	not active	not active
S. fruticosa	55.2±1.0	1.08±0.02	3.03±0.13
S. crypthantha	55.1±3.3	1.09±0.06	2.55±0.14
S. sclarea	53.1±3.4	not active	not active
S. palaestina	50.4±2.9	not active	not active
BHT	62.6±5.3	1.16±0.04	2.13±0.12

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[1]Percentage inhibition (capacity to inhibit peroxide formation in linoleic acid).

[2]Induction index: induction time of sunflower oil sample/induction time of sunflower oil.

[3]Results are expressed as mean ± standard deviation (n = 3).

Not active: (≤1%). The samples were not active in the peroxidation of sunflower oil).

The formation of peroxidation products was inhibited by the addition of crude extracts in a dose-dependent manner. The extracts of *S. halophila*, *S. sclarea*, *S. cilicica*, and *S. palaestina* were not active in the peroxidation of sunflower oil. *S. fruticosa*, *S. crypthantha*, *S. chrysophylla*, and *S. tomentosa* showed slight activity at a level of 0.02%. Their effects were dominant at a concentration of 1%. Among the extracts, only *S. fruticosa* and *S. crypthantha* showed higher antioxidant power than that of BHT.

Free Radical Scavenging Activity on DPPH. Free radical scavenging effects of the fractions on DPPH were estimated according to the method of Sanchez-Moreno [15]. The absorbance of samples was measured spectrophotometrically at 517 nm. *S. chrysophylla*, *S. halophila*, *S. tomentosa*, and *S. fruticosa* exhibited free radical scavenging activity (Fig. 1). Their activities were comparable with that of BHT at all the concentrations tested. Free radical scavenging activities were in the following order: *S. chrysophylla* > BHT > *S. halophila* > *S. tomentosa* > *S. fruticosa* > *S. crypthantha* > *S. cilicica* > *S. sclarea* > *S. palaestina*.

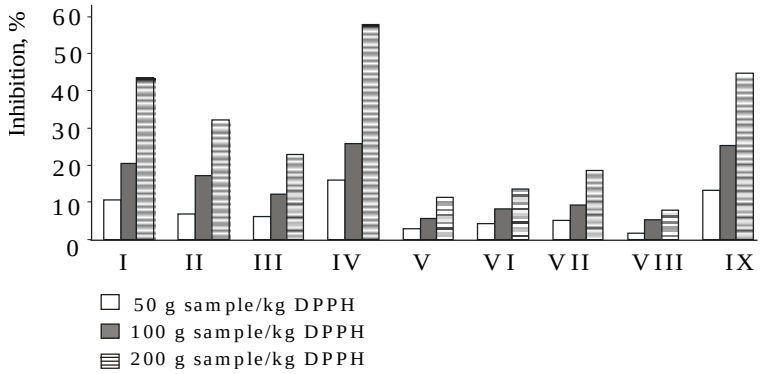

Fig. 1. Free radical scavenging effects of *Salvia* species on DPPH.
I - *S. halophila*, II - *S. tomentosa*, III - *S. fruticosa*, IV - *S. chrysophylla*, V - *S. sclarea*, VI - *S. cilicica*, VII - *S. crypthantha*, VIII - *S. palaestina*, IX - BHT.

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