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ANTIOXIDANT ACTIVITY AND TOTAL PHENOLIC AND FLAVONOID CONTENTS OF *SALVIA AMPLEXICAULIS* LAM. EXTRACTS

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Abstract - This study was designed to examine the *in vitro* antioxidant DPPH free radical-scavenging activity, and the total phenolic and flavonoid content of ethanol and methanol extracts of *Salvia amplexicaulis* Lam. in the whole plant and different parts, leaves, stems and flowers. The largest amounts of extract yield were obtained from the flowers, 14.14% and 12.00 % (w/w) in the ethanol of methanol extracts, respectively. The ethanol extract of leaves (16.07 μg/ml) and methanol extract of the whole plant (21.28 μg/ml) showed the highest activity against the DPPH radical. The ethanol extract of the leaves was the richest in phenols (222.40 mg GAE/g) and flavonoids (49.81 mg QE/g), whereas the methanol extract of the whole plant contained the highest amount of phenolics (180.89 mg GAE/g) and flavonoids (38.15 mg QE/g). A very strong linear correlation between antioxidant activity and the phenolic content of the extracts was established. The obtained results suggest that *S. amplexicaulis* could be regarded as an important source of natural antioxidants.

Keywords: *Salvia amplexicaulis*, Lamiaceae, DPPH, phenolics, flavonoids, extracts

INTRODUCTION

Since ancient times, aromatic plants have been used for improving the flavor and organoleptic properties of food, as well as for the treatment of various diseases. Studies have revealed a wide range of biological activities, including antioxidant, antimicrobial, antiviral, antitumor effects of different aromatic plants and shown them to be useful in food and cosmetics preservation, and as valuable ingredients in the foods that combine nutritional and medicinal benefits, so-called functional foods.

Oxidative cell damage assumes an important place in the etiology of many diseases such as atherosclerosis, arthritis, cardiovascular disorders, Alzheimer's disease, and cancer (Halliwell, 1994, 1995, 1999). Free radicals are produced in the human body from normal metabolism or induced by physical and/or chemical factors in the environment. Harmful effects can be decreased by the intake of antioxidant substances. Antioxidants work by donating an electron to a molecule that has been compromised by oxidation, bringing it back into a state of proper function (Lu and Foo, 2002). Synthetic antioxidants,

such as butylated hydroxyanisole (*BHA*) and butylated hydroxytoluene (BHT) are very effective, but they can induce tumors at high doses and after long-term treatment (Kahl and Kappus, 1993). For this reason, there is increasing interest for naturally occurring antioxidants that can be used in food, cosmetic and pharmaceutical products to replace synthetic antioxidants. Numerous studies have demonstrated that medicinal plants contain various components possessing antioxidant properties with beneficial health effects.

Among natural antioxidants, phenolic substances have been of special interest because they are widely distributed in the plant kingdom, constituting one of the main classes of secondary metabolites, with more than 8 000 phenolic structures currently known, ranging from simple molecules, such as phenolic acids, to highly polymerized substances such as tannins (Dai and Mumper, 2010). Phenolic compounds are responsible for the major organoleptic characteristics of plant-derived foods and beverages, particularly color and taste properties, and they contribute to the nutritional qualities of fruits and vegetables (Tapas et al., 2008). Among these compounds, the flavonoids constitute one of the most ubiquitous groups of all plant phenolics. Several thousand different naturally occurring flavonoids have been discovered. Flavonoids are present in a wide variety of edible plant sources, such as fruits, vegetables, nuts, seeds, grains, tea and wine (Marin et al., 2003). Flavonoids are the major active nutraceutical ingredients with health benefits. They are recognized to possess anti-inflammatory, anti-allergic, hepatoprotective, antithrombotic, antiviral and anticarcinogenic activities (Tapas et al., 2008).

The genus *Salvia* (Lamiaceae) represents an enormous and cosmopolitan assemblage of nearly 1 000 species displaying a remarkable range of variation (Walker et al., 2004). Several *Salvia* species, commonly called sages, are cultivated worldwide for use in traditional medicine and for culinary purposes.

Like other members of the Lamiaceae family, *Salvia* species show diverse biological activities manifested by the different constituents of essential oil and/or extracts. These are antioxidant (Tepe et al., 2006), antiproliferative (Fiore et al., 2006), anticholinesterase (Orhan et al., 2012, 2013), antimicrobial (Petrović et al., 2009), antiviral, anticancer, anti-inflammatory, cardio- and neuroprotective activities (Perry et al., 2003; Wang, 2010). Numerous chemical studies have revealed that *Salvia* species are a rich source of polyphenols, with an excess of 160 polyphenols identified, some of which are unique to the genus (Lu and Foo, 2002).

Salvia amplexicaulis Lam. is a perennial plant with a height of up to 80 cm. Its leaves are short petiolate or sessile, densely eglandular – pubescent below and subglabrous above. The flowers have a pubescent calyx and violet corolla; 6-8 flowers are grouped in the inflorescences. This species is distributed on the Balkan Peninsula (Hedge, 1972).

S. amplexicaulis was partially investigated previously. Petrović et al. (2009) analyzed the essential oil composition and antimicrobial activity of the oil. They identified fifty-one compounds and found that the essential oil contained a high amount of sesquiterpenes (81.1%) with germacrene D, viridiflorol, caryophyllene oxide and ß-caryophyllene as the main components. The microbial growth inhibitory effect was more pronounced on Gram-positive bacteria and *Candida albicans* compared to Gram-negative bacteria. Ulubelen (2003) proved the vasodepressor effect of several diterpenoids, steroids and crude extract of *S. amplexicaulis* that were isolated from its roots (Kolak et al., 2001). Orhan et al. (2012) screened the ethyl acetate and methanol extracts of 16 Turkish *Salvia* species for their inhibitory activity against the enzymes linked to neurodegeneration – acetylcholinesterase, butyrylcholinesterase, lipoxygenase and tyrosinase, and tested their antioxidant activity using DPPH and FRAP assays. They found that *Salvia amplexicaulis* extracts possessed antioxidant and neurobiological activities, with higher activities obtained for the methanol extract.

Several studies examined the various parts of medicinal plants for the antioxidant and antimicrobial

activities of essential oils and/or extracts (Veličković, 2003; Wannes et al., 2010; Baňo et al., 2003; Šamec et al., 2010; Končić et al., 2010; Stanković et al., 2010, 2011; Barros et al., 2010; Rafat et al., 2010; Siddique et al., 2010; Riahi et al., 2013). The authors found differences in the chemical composition and biological activities among the plant parts examined.

The aim of the present study was to evaluate the radical-scavenging activities of ethanol and methanol extracts of *S. amplexicaulis* plant parts by 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging assay. Because of the important roles of phenolics and flavonoids as antioxidants, the amounts of total phenolics and total flavonoids of whole plant, stem, leaves and flower ethanol and methanol extracts were also determined.

MATERIALS AND METHODS

Chemicals

Methanol, ethanol and distillated water were purchased from Zorka Pharma (Šabac, Serbia). Gallic acid, quercetin, ascorbic acid, 2(3)-t-Butyl-4-hydroxyanisole (BHA), 3,5-di-tert-butyl-4-hydroxytoluene (BHT), 2,2-dyphenyl-1-picrylhydrazyl (DPPH), potassium acetate ($C_2H_3KO_2$), sodium carbonate anhydrous (Na_2CO_3), aluminum nitrate nonahydrate ($Al(NO_3)_3$ x $9H_2O$) and Folin Ciocalteu's phenol reagent were obtained from Sigma Chemicals Co. (St Louis, MO, USA). All chemicals used were of analytical grade purity.

Plant material

Aerial parts of investigated plant *Salvia amplexicaulis* Lam. were collected at the end of flowering stage in July 2011 from Pletvar locality, Macedonia. Plant material was dried and kept in the shade at room temperature until further processing. A voucher specimen was deposited in the Herbarium of the Institute of Botany and Botanical Garden "Jevremovac" (BEOU; voucher No. 16673).

Preparation of plant extracts

Extracts of whole plants were prepared, as well as from different parts of the plants. Five g of plant material (whole plants, leaves, stems and flowers, mainly calyces and pedicels) were ground into small pieces (2-6 mm) in a cylindrical crusher and extracted with 50 ml 96% ethanol by a classic maceration procedure during 24 h at room temperature (10% w/v). The mixture was exposed to ultrasound 1 h before and after 24 h maceration, the extracts were filtered (Whatman No. 1) and evaporated under reduced pressure with a rotary evaporator (Buchi Rotavapor R-114). After evaporation of the solvent, the obtained crude extracts (Table 1) were stored at +4°C.

Evaluation of DPPH scavenging activity

For evaluation of the antioxidant activity of extracts, the 2,2-dyphenyl-1-picrylhydrazyl (DPPH) free radical scavenging method (Blois, 1958) with slight modifications was used. This assay is spectrophotometric and uses stable the DPPH radical as reagent. Stock solutions of dry extracts were prepared in 96% ethanol/methanol in a concentration of 1 000 μg/ml (w/v). Stock extract solution was diluted with the methanolic solution of DPPH (40 μg/ml) to adjust the final volume of the reaction mixture (4 000 μl) of the test tube (extract concentrations 10, 15, 20, 25, 30 μg/ml (v/v)). Methanol was used as a blank, while methanol with DPPH solution was used as a control. BHA, BHT and ascorbic acid were used as positive controls (standards). All samples were measured in triplicate. The absorbance was measured at 517 nm after 30 min in the dark at room temperature, using a JENWAY 6305UV/Vis spectrophotometer. The decrease of absorption of DPPH was calculated as follows:

$$\text{Inhibition of DPPH radical (\%)} = [(A_C - A_S)/A_C] * 100\%$$

where A_c is the absorbance of the control, A_s is the absorbance of the test samples. IC_{50} values (μg/ml) (concentrations of the test samples and standard antioxidants that provided 50% inhibition of the DPPH radical) were calculated from the DPPH absorption curve at 517 nm.

Determination of the total phenolic content

The total phenolic content of *Salvia amplexicaulis* extracts was performed by spectrophotometry (Singleton and Rossi, 1965). The reaction mixture was prepared by mixing 0.2 ml of the ethanol/methanol extract (1 mg/ml) with 1 ml of 10% Folin & Ciocalteu's reagent dissolved in water. After 6 min, 0.8 ml of 7.5% Na_2CO_3 was added. The blank contained distillated water instead of the extract. Absorbance was recorded at 740 nm after 2 h incubation at room temperature. The same procedure was repeated for the standard solution of gallic acid (GAE). The phenolic content in the samples was calculated from the standard curve and expressed as mg GAE/g dry extract, averaged from three measurements.

Determination of flavonoid concentration

Flavonoid concentrations of samples were measured spectrophotometrically according to the procedure of Park et al. (1997). The reaction mixture was prepared by mixing 1 ml of ethanol/methanol extract solution (1 mg/ml) with 4.1 ml of 80% ethanol, 0.1 ml of 10% $Al(NO_3)_3x9H_2O$, and 0.1 ml 1 M CH_3COOK. The blank contained 96% ethanol instead of the extract. After 40 min of incubation at room temperature, absorbance was measured at 415 nm. The same procedure was repeated for the 96% ethanol solution of antioxidant quercetin (QE) in order to construct a calibration curve. The concentration of flavonoids in the samples (mg/ml) was expressed as mg QE/g dry extract, averaged from three measurements.

Statistical analysis

All measurements were carried out in triplicate and expressed as the average of three measurements ± standard deviation. Calculations and construction of curves were performed using MS Office Excel, 2007.

RESULTS AND DISCUSSION

Recent developments in biomedical science emphasize the involvement of free radicals in many diseases (Halliwell, 1999, 2009; Griendling and FitzGerald, 2003; Valko et al., 2006). Dietary antioxidant intake may be an important strategy for inhibiting or delaying the oxidation of susceptible cellular substrates, and is thus relevant in disease prevention. Phenolic compounds such as flavonoids, phenolic acids, diterpenes and tannins have received attention for their high antioxidative activity. Crude extracts of herbs and spices, and other plant materials rich in phenolics, are of increasing interest in the food industry because they retard oxidative degradation of lipids and thereby improve the quality and nutritional value of food (Shuka et al., 2009). Phytochemical investigations have shown that *Salvia* species are mainly rich in polyphenolics and flavonoids, which exhibit antioxidant properties (Božin et al., 2007).

Table 1. Yields of ethanolic and methanolic extracts of *Salvia amplexicaulis*

Type of extract	% yield of ethanol extract (w/w)	% yield of methanol extract (w/w)
Whole plant	13,18	8,78
Leaves	10,18	8,16
Stems	7,52	5,92
Flowers	14,14	12,00

In this work, total phenolic and flavonoid content, and antioxidant activity *in vitro* were determined and compared for ethanol and methanol extracts of whole *S. amplexicaulis* plant, as well as for leaves, stems and flowers, separately. The yield of extracts obtained from 5 g of different plant parts and the whole plant of *S. amplexicaulis*, using different solvents, is listed in Table 1. The yields of extracts varied depending on the plant part. The largest yields were obtained from the flowers and whole plant, 14.14 and 13.18% (w/w) for the ethanol, and 12.00 and 8.78% (w/w) for the methanol extracts, respectively. The ethanol extracts showed a higher yield than the methanol extracts. Veličković et al. (2003) obtained lower yields of *Salvia officinalis* dry ethanol extracts: 3.5% (flowers); 3.1% (leaves), 1.2% (stem). Fiore et al. (2006) found that the yields of the methanol extracts of six *Salvia* species from Jordan ranged from 118 to 183 mg/g,

Table 2. Total phenol and flavonoid content of ethanol and methanol extracts of different *Salvia amplexicaulis* parts expressed in terms of GAE equivalents (mg GAE/g dry extract) for total phenols and QE equivalents (mg Q/g dry extract) for flavonoids. Values are presented as mean ± standard deviation.

Salvia amplexicaulis extracts	Ethanol extracts		Methanol extracts	
	Total phenols (mg GAE/g dry extract)	Total flavonoids (mg QE/g dry extract)	Total phenols (mg GAE/g dry extract)	Total flavonoids (mg QE/g dry extract)
Whole plant	138.05 ± 0.997	27.35 ± 0.601	180.89 ± 1.708	38.15 ± 0.967
Leaves	222.40 ± 0.643	49.81 ± 0.820	154.33 ± 0.845	33.69 ± 0.460
Stems	173.67 ± 1.273	28.77 ± 0.577	124.31 ± 1.16	15.46 ± 0.920
Flowers	112.42 ± 0.601	21.85 ± 1.846	140.31 ± 0.370	20.59 ± 1.350

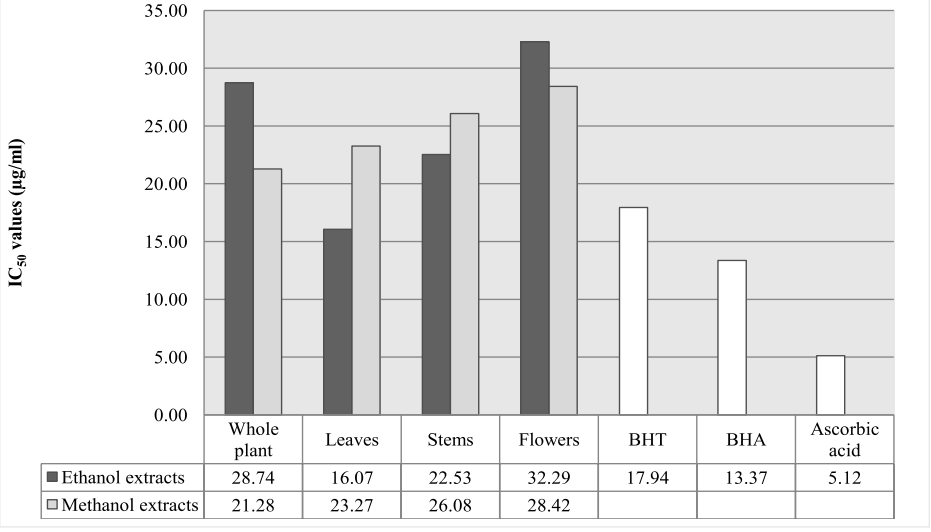

Fig. 1. DPPH scavenging activity of ethanol and methanol extracts of *Salvia amplexicaulis* presented as IC_{50} values (µg/ml).

with the highest amount of extract obtained from leaves and aerial plant parts. The yield of the methanolic extract of the aerial parts of 16 *Salvia* species from Turkey ranged from 3.05% for *S. amplexicaulis,* to 66.17% for the endemic *S. ekimiana* (Orhan et al., 2012). Senol et al. (2010) obtained yields of the methanolic extract of *Salvia* taxa from 2.88-13.41%. Stanković et al. (2010) obtained similar yield ratios of methanol extracts of different *Teucrium chamaedrys* L. var. *glanduliferum* plant parts (from 1.04% for stems, to 2.44% for leaves). Our results show that the extract yields vary depending on plant part and solvent used for the extraction.

Besides variations in extract yield, the variations in essential oil yield and quality depend on the part of the plant, season, growth phase, localities (Perry et al., 1999; Chalchat and Ozkan, 2008; Zaouali et al., 2013).

DPPH is a stable free radical. When a solution of DPPH is mixed with a substance that can donate a hydrogen atom, this gives rise to the reduced form with the loss of the violet color (Molyneux, 2004). DPPH scavenging activity was measured as the percentage of decreased DPPH radical adsorption. Concentrations of the test samples and standard antioxidants providing 50% inhibition of DPPH radicals (IC_{50}; µg/ml) were calculated from the absorption curve at 517 nm. IC_{50} values of the ethanol and methanol extracts of whole plants and different parts of *S. amplexicaulis* are presented in Fig. 1. As can be seen, the leaf

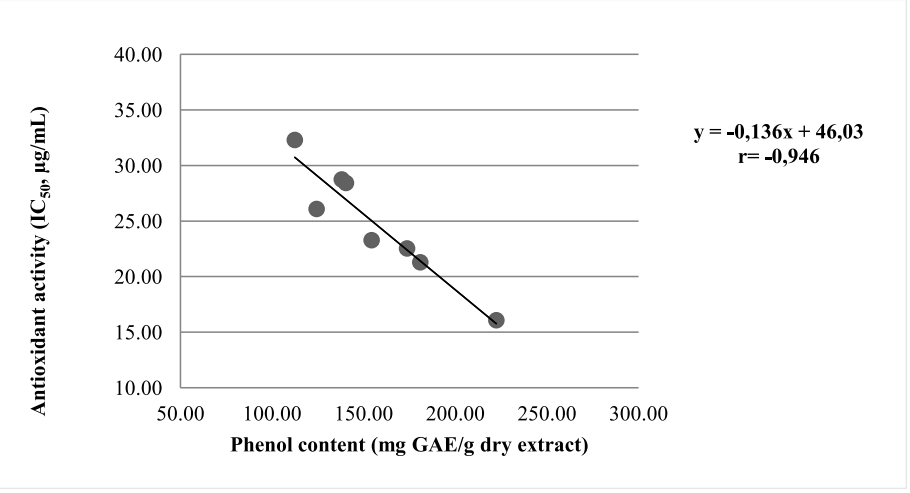

Fig. 2. Linear correlation between the amount of total phenols and antioxidant activity. Coefficient of determination R^2 = 0,894 coefficient of correlation r= -0,946. Correlation is significant at the 0.05 level.

ethanol extract had the strongest activity against the DPPH radical (16.07 µg/ml), followed by the stems, whole plants and flowers (22.53, 28.74, 32.29 µg/ml, respectively). The methanolic extract of the whole plant was the most potent in DPPH decolorization (21.28 µg/ml), followed by leaves (23.27 µg/ml), stems (26.08 µg/ml) and flowers (28.42 µg/ml). The IC_{50} values of the synthetic antioxidants BHT, BHA and ascorbic acid were 17.94, 13.37 and 5.12 µg/ml, respectively. Ethanolic extracts of leaves and stems showed stronger antioxidant activity compared to the methanolic extracts.

Kamatou et al. (2010) have measured a wide range of IC_{50} values for methanol:chloroform (1:1) extracts of 16 South African *Salvia* species (from 1.6 to 74.5 µg/ml using DPPH). Tosun et al. (2009) found that the IC_{50} values of methanol extracts of eight *Salvia* species from Turkey ranged from 18.3-88.2 µg/ml. In our experiments, the ethanol extract of leaves showed a noticeably strong DPPH activity (16.07 µg/ml), stronger than that of the synthetic antioxidant BHT (17.94 µg/ml) and similar to BHA (13.37 µg/ml), which are used to prevent fats and oils in food from becoming rancid.

When the comparisons of different extraction solvents for different plant parts were done, the results varied. Stanković et al. (2011) studied the total phenolic content, flavonoids and antioxidant activity of different extracts of *Teucrium montanum* L. var. *montanum* whole plant, leaves, stems and flowers and obtained the best result for the water extract of the whole plant in DPPH assay. For *Teucrium chamaedrys* L. var. *glanduliferum*, the highest capacity to neutralize DPPH radicals was found in the methanol extract of the stem (Stanković et al., 2010). Comparing the antioxidant capacity of flower and leaf infusions of *Teucrium arduini* L. from different localities, Šamec et al. (2010) obtained better results for the leaf infusion. Rafat et al. (2010) obtained the highest antioxidant potential for *Andrographis paniculata* leaf ethanolic extracts in two tests, followed by stem and fruit extracts, but the fruit extract exhibited the highest DPPH scavenging activity. Leaf methanolic extracts of *Aegle marmelos* (Siddique et al., 2010), *Myrtus communis* var. *italica* (Wannes et al., 2010), *Artemisia absinthium* (Riahi et al., 2013) and *Malva sylvestris* (Barros et al., 2010) showed the highest antioxidant capacity compared to other plant parts. The aqueous leaf extracts of *Moltkia petraea* demonstrated superior antioxidant activity in most of the applied assays compared to stems and flowers (Končić et al., 2010). Most results obtained on the other plants showed the best antioxidant effect for leaf extracts, as was proved for *S. amplexicaulis*

ethanol extract in our investigation, which is probably connected to the glandular trichomes distribution on the plant and the secondary metabolites produced in them, such as phenolic compounds, which are potent antioxidants.

The quantities of phenols and flavonoids in the examined plant extracts are presented in Table 2. The whole plant and all three examined parts of *S. amplexicaulis* had significantly different contents of phenolics. Total phenol content is expressed as gallic acid equivalents (mg GAE/g dry extract). The ethanol extract of leaves was the richest in phenols (222.40 mg GAE/g), while the flowers contained almost half the amount (112.42 mg GAE/g). The whole plant and stems were moderately abundant in phenolic compounds (138.05 and 173.67 mg GAE/g, respectively). The methanol extract of the whole plant (180.89 mg GAE/g) and leaves (154.33 mg GAE/g) contained the highest quantity of phenolics, followed by flowers (140.31 mg GAE/g) and stems (124.31 mg GAE/g).

The ethanol extracts of leaves and stems were richer in phenolic compounds than in methanol extracts. In the case of the whole plant and flowers, the methanol extracts contained more phenolics.

Stanković et al. (2011) have made a comparative analysis of phenol content in different plant parts using different solvents and concluded that the highest concentration of phenolic compounds in extracts were obtained using solvents of high polarity. In water and methanolic extracts, the highest concentration of phenolic compounds was obtained in the leaves. Other authors who analyzed the quantitative and qualitative phytochemical characteristics of plant organs obtained different values for the leaf, stem and flower. Bystricka et al. (2010) reported that the concentration and dynamics of polyphenol synthesis in plant organs depend on the plant cultivar, studied organ and growth phase, and they obtained the best results for the flowers and leaves of *Fagopyrum esculentum,* depending on the growth phase. Rafat et al. (2010) obtained a higher concentration of phenolic compounds in the leaf ethanolic extract of *Andrographis paniculata* compared to the stem extract, as was obtained in our research. The leaf methanol extracts of *Aegle marmelos* (Siddique et al., 2010), *Myrtus communis* var. *italica* (Wannes et al., 2010), *Artemisia absinthium* (Riahi et al., 2013) and *Malva sylvestris* (Barros et al., 2010) were also found to be richest in phenols than the other plant parts. In our research, a higher phenol content in *S. amplexicaulis* was found compared to the phenol content found in methanol extracts of eight *Salvia* species collected in Turkey, where it varied from 50.3 to 101.2 mg GA/g (Tosun et al., 2009). Our results are in agreement with the range of values (45-211 mg GAE/g) specified previously for total phenol methanol/chloroform extracts of sixteen South African *Salvia* species (Kamatou et al., 2010). The main components of the investigated extracts were rosmarinic acid, carnosic acid, carnosol and ursolic acid (Kamatou et al. 2010). The total phenol content of ethanol extract for 14 Turkish *Salvia* species ranged from 57.10-218.09 mg GAE/g and 8.29-108.78 mg QE/g for flavonoids (Orhan et al., 2013).

Flavonoid concentrations are expressed as quercetin equivalents (mg QE/g dry extract) and ranged from 49.81 mg Q/g) in leaf, to 21.85 mg QE/g in flower ethanol extracts. Methanol extracts contained fewer flavonoids than the ethanol extracts, except the whole plant methanol extract (38.15 mg Q/g), followed by leaves (33.69 mg Q/g), stems (20.59 mg Q/g) and flowers (15.46 mg Q/g). The concentration of flavonoids in plant parts differed greatly from the value obtained for the whole plant, as was found for *Teucrium chamaedrys* L. var. *glanduliferum* (Stanković et al., 2010) and *Aegle marmelos* (Siddiqui, 2010). The highest concentration of flavonoids in leaf extracts was also obtained in other studies (Siddique et al., 2010; Stanković et al., 2010; Rafat et al., 2010; Barros et al., 2010; Riahi et al., 2013). Different results have reported that for *Myrtus communis* var. *italica* the highest content of total flavonoids was observed in stem methanol extract (Wannes et al., 2010), and for *Euphorbia helioscopia*, the highest flavonoid content, as well as phenols, followed by the highest antioxidative activity, were found in flower methanolic extracts (Maoulainine et al., 2012).

The positive correlation between antioxidant activity and total phenol content of extracts is established in *S. amplexicaulis* (Fig. 2). It can be noticed that an increase in the phenol content of extract decreases the IC_{50} value, i.e. increases their scavenging DPPH free-radical activity (negative correlation, r = -0.946). Phenols are very important plant constituents because of their scavenging ability on free radicals due to their hydroxyl groups. Therefore, the phenolic content of plants may contribute directly to the antioxidant activity of extracts (Tosun et al., 2009). Plants belonging to the Lamiaceae family are very rich in polyphenolic compounds. The major phenolic compounds identified in the extracts of sage are rosmarinic acid, carnosic acid, salvianolic acid and its derivates carnosol, rosmanol, epirosmanol, rosmadial and methyl carnosate (Madsen and Bertelsen, 1995; Lu and Foo, 2001). Among these, rosmanol is a major constituent of many *Salvia* species and possesses strong antioxidant capacity because these groups cause phenols to more easily donate hydrogen atoms to activate free radicals to interrupt the chain reaction of antioxidation (Weng and Wang, 2000). The DPPH scavenging capacity of these extracts may be mostly related to their phenolic hydroxyl groups.

A positive linear correlation between the total content of phenolic compounds and the antioxidant activities for aqueous and methanolic extracts of 112 Chinese medicinal plants (Cai et al., 2004), as well as various Jordanian plants (Alali, 2007), has been established. A positive linear correlation was observed between the total phenolic content and antioxidant activity of the methanolic extracts of eight Turkish *Salvia* species (Tosun et al., 2009). Three different extracts of *Salvia sclarea, S. glutinosa,* and *S. pratensis* were analyzed by Miliauskas et al. (2004) and a correlation between the antioxidant capacity of extracts and total phenolic content was established.

In a number of studies of the differences in antioxidant activities of plant parts, correlations between the radical-scavenging capacities of examined extracts with total phenolic compound content was observed. For *Andrographis paniculata*, the positive correlation between free radical-scavenging capacity and the content of phenolic compounds was found in the fruit, leaf and stem extracts of the plant (Rafat et al., 2010). The methanolic extracts of plant parts of *Artemisia absinthium* (Riahi et al., 2013), *Aegle marmelos* (Siddiqui et al., 2010), *Myrtus communis* (Wannes et al., 2010), *Malva sylvestris* (Barros et al., 2010) and different *Teucrium* species (Stanković et al., 2010, 2011) showed significant linear correlation between the values of the concentration of phenols and antioxidant activity. Siddiqui et al. (2010) concluded that a greater amount of phenolic compounds leads to more powerful radical-scavenging effect.

CONCLUSION

Methanol and especially ethanol extracts obtained from the whole plant and separate parts of *S. amplexicaulis* showed very strong *in vitro* antioxidative activity against the DPPH free radical, similar to the reference antioxidants (BHA and BHT). All tested extracts, especially those of the leaves, exhibited a high content of phenols and flavonoids. Comparison of the antioxidant activities with the phenolic contents suggest that these compounds are responsible for the antioxidant activity. The results of this study revealed the variability and importance of separate examination of the phenol composition and antioxidative potential of plant parts in comparison to the whole plant,. Our results suggest that the herb *S. amplexicaulis* is a rich source of antioxidants, with potential application in the protection and preservation of certain foods and nutraceuticals.

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