



## The comparative investigation of the antioxidant activities of some species belonging to the Lamiaceae and Poaceae families

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### Lamiaceae ve Poaceae familyalarına ait bazı türlerin antioksidan savunma kapasitelerinin karşılaştırmalı olarak incelenmesi

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**Abstract:** The antioxidant compounds of plants have widely been investigated for the purpose of medical and industrial uses due to their aroma, color, smell and protective properties. In the study, 16 species from Lamiaceae and 14 species from Poaceae were collected from the Northeastern Mediterranean region and then they were analysed. The dry weight and water conditions of the plant samples were identified. It was identified that there wasn't a significant difference between these two families in respect to their values of chlorophyll and carotene. The mean xanthophyll content was higher in the Lamiaceae species (236 µg g<sup>-1</sup> FW) than in the Poaceae species (142 µg g<sup>-1</sup> FW) and total antioxidative capacity was higher in Lamiaceae species (5.19 mg g<sup>-1</sup> FW) than Poaceae species (3.49 mg g<sup>-1</sup> FW). On the other hand, mean soluble phenolics were measured as 1.83 mg g<sup>-1</sup> FW in Poaceae species and 1.67 mg g<sup>-1</sup> FW in Lamiaceae species. Superoxide dismutase enzyme activity was higher in Poaceae species. The findings revealed that significant differences could exist among the families and further comparative studies should be performed for the determination of the biochemical resources.

**Key words:** carotenoids; Mediterranean vegetation; phenolics; pigments; superoxide dismutase

**Özet:** Bitkilerin antioksidan bileşikleri aroma, renk, koku ve koruyucu özellikleri nedeniyle tıbbi ve endüstriyel kullanım amacıyla yoğun olarak araştırılmaktadır. Bu çalışmada, Lamiaceae'den 16 tür ve Poaceae'den 14 tür, Kuzeydoğu Akdeniz bölgesinden toplandı ve analiz edildi. Bitki örneklerinin kuru ağırlık ve su durumları belirlendi. Bu iki familyanın üyeleri arasında klorofil ve karoten değerleri bakımından önemli bir fark olmadığı belirlendi. Lamiaceae türlerinde ortalama ksantofil içeriği (236 µg g<sup>-1</sup> FW) Poaceae türlerinden (142 µg g<sup>-1</sup> FW) ve total antioksidatif kapasite Lamiaceae türlerinde (5.19 mg g<sup>-1</sup> FW) Poaceae türlerinden (3.49 mg g<sup>-1</sup> FW) daha yüksek bulundu. Buna karşın toplam çözünür fenolikler Poaceae türlerinde ortalama 1.83 mg g<sup>-1</sup> FW, Lamiaceae türlerinde 1.67 mg g<sup>-1</sup> FW olarak ölçüldü. Süperoksit dismutaz enzim aktivitesi Poaceae türlerinde daha yüksek bulundu. Bulgular iki familya arasında önemli farklılıklar bulunabileceğini ve biyokimyasal kaynakların belirlenmesi bakımından karşılaştırmalı çalışmaların çoğaltılması gerektiğini ortaya koymaktadır.

**Anahtar Kelimeler:** Akdeniz vejetasyonu, fenolikler, karotenoidler, pigmentler, süperoksit dismutaz

### 1. Introduction

Biological systems have been protected from the destructive effects of oxygen by antioxidant defense mechanisms (Alscher and Hess, 1993). The mechanisms of tolerance or avoidance of oxidative stress differ among plant groups (Cai et al., 2004). The analysis of oxidative defense components can make a contribution for the determination of the differences among plant families. The primary elements serving for oxidative protection in plants are phenolic compounds of which antioxidants effects have been known and which have large diversity (Wojdylo et al., 2007; Rice-Evans et al., 1996). The strong water-soluble antioxidants such as glutathione and ascorbic acid are quite common in plant families. The levels of the oil-soluble antioxidants such as carotenoids and tocopherol differ among plant groups as well. The similar differences could be observed in antioxidant enzyme activities such as SOD, GR, catalase and peroxidases (Oncel et al., 2004).

Increasing reactive oxygen species in plants under stress are the harmful oxygen forms with higher chemical reactivity as compared with the oxygen molecules (Van Breusegem and Dat, 2006). Free radicals are high-powered, unstable compounds containing the electrons that not having constituted one or more pairs, in their outer atomic orbitals. Free radicals cause a damage by

receiving electrons from many biological materials such as proteins, lipids, nucleic acids and coenzymes (Mittler, 2002). The studies evaluating the plant families with different biochemical characteristics in respect to their antioxidant capacities, reported significant differences (Cai et al., 2004; Wojdylo et al., 2007). There are evidences implying that the oxidative stress protection mechanisms of the Lamiaceae and Poaceae families, both of which have different biochemical characteristics, are also different (Oncel et al., 2004).

Lamiaceae's family has been represented with approximately 250 genus and 7000 species in the world (Kahraman et al., 2009). Members of this family range intensively in Mediterranean countries primarily, South West Asia and South America. Turkey is one of the important gene centers of Lamiaceae. This family has been represented with 574 species within 45 genus in Turkey (Güner et al., 2000). Most of the members of the Lamiaceae have a great importance in the fields of medical, pharmaceutical, food, cosmetic and perfumery because of being rich in essential oils, aromatic compounds, secondary metabolites (Başer, 1993). On the other hand, the use of ethnobotany is quite common among the members of the family (Matkowski et al., 2008; De Marino et al., 2012). Because of their high

polyphenol content, Lamiaceae's plants were investigated as natural antioxidant resources (Rice-Evans et al., 1996).

The members of the Poaceae's family usually in the form of annual or perennial herbaceous plant, rarely in the form of bush or tree. Their roots are in the type of fibrous root and they contain some rhizome. Their stem is vertical, ascendant, leaning and creeping. Since their stem doesn't grow in width, the inner of their stem is empty, excluding nodiums. Their seeds are rich in starch. This cosmopolite family contains about 650 genus and species of much more than 9000. It has 512 species belonging to 142 genus in Turkey (Guner, 2000). It's many genus have economical value as being cultivated. In addition, it's species with sugar and oil contents are also available. They are feed and food plants which are rich in antioxidant phenolic acids and xylooligosaccharides (Reddy and Krishnan, 2013).

The species belonging to the Poaceae and Lamiaceae have been the subject of many studies in respect to their antioxidative capacities (Cai et al., 2004; Markowskaya et al., 2012). In this study, it was intended to perform comparative investigation of the antioxidative capacities of Poaceae and Lamiaceae families and determination of whether any difference exist in respect to their antioxidative capacities. The findings of this study can make a useful contribution for the determination of the systematical, physiological and biochemical characteristics of both families. Lamiaceae and Poaceae's species, which exist commonly in the Eastern Mediterranean region, were collected from different localities and analysed in laboratory in respect to their content of antioxidant featured substances and enzymes. It should not purely be a review of the subject area, and should not contain the findings or the conclusions.

## 2. Material and Methods

### 1.1. Plants

The plants which are the subject of the study were collected in July and August 2011 from the Çamlıyayla (Göpter-Çuvalğı), Kazanlı, Apsun, Bükdeğirmeni, Kuyuluk, Aydınçık, Değirmençay, Işıklı ve Gözne districts, which situated within the borders of Mersin province. This area is located in C4 square according to the grid system specified in Turkey's flora. 16 of the species of 30 plant investigated belong to the Lamiaceae, 14 of them belong to the Poaceae. Only fresh leaves of Lamiaceae's samples, both leaves and stems of Poaceae's samples were cut out and stored in a freezer. The samples were taken from three different region for each species, the results of the analysis were given in average values of these samples.

### 1.2. Dry Weight and Relative Water Content (RWC)

Leaf dry matter content were determined as the difference between fresh weight and dry weight after drying at 110°C for 24 hours in an oven. Measurements of relative water content (RWC) were taken from segments of leaves. After floating on distilled water that allowed the leaf segments to rehydrate for 2 h at 20°C, they were blotted dry and weight. The same segments were dried overnight at 110°C and weight again. RWC of leaves was calculated according to formula:  $RWC = 100 \times [(fw-dw) / (turgid w-dw)]$ .

### 1.3. Chlorophyll Content

Chlorophyll extraction from fresh leaf material was carried out with 80% acetone (buffered to pH 7.8 with phosphate buffer). The chlorophyll a, chlorophyll b and total chlorophyll measurements were done with spectrophotometer. Chlorophyll contents were calculated according to Porra et al. (1989) and chlorophyll a/b ratios were determined.

### 1.4. Carotenoid Content

Fresh leaf material (0.5 g) were ground in pre-chilled mortar in 5 ml acetone containing 200 mg Na<sub>2</sub>SO<sub>4</sub> and then filtered through glass fiber disks (Whatmann GF/A). The volume of the acetone extracts was reduced in rotary evaporator and then resuspended in 1 ml chloroform. Fifty microliters of the extracts and standarts were applied to silica gel TLC plates (20x20, 0.25 mm thickness). The chromatograms were developed with hexane, diethyl eter, acetone, 60:30:20, v:v:v (Moore, 1974). Xanthophyll and  $\beta$ -caroten spots were scraped from the TLC plates and centrifuged in 5 ml acetone for 5 min at 5000g. The absorbance of supernatants was determined at a wavelength of 450 nm by a spectrofootometer, against  $\beta$ -caroten and lutein standarts.

### 1.5. Total Antioxidant Capacity

0.5 g plant sample was crushed in porcelain mortar with 5 ml methanol (96%) for determination of total antioxidative capacity. The extract was centrifuged for 5 minutes at 5000 g and supernatant was taken. A reactive containing 6 M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate was prepared. 150  $\mu$ l supernatant was mixed with the reactive in a test tube so that last volume would be 3 ml. The tubes was maintained at 95 °C for 90 min. and then cooled until room temperature and their absorbances were measured at 695 nm. Total antioxidative capacity was calculated as the equivalent of ascorbic acid (Prieto et al., 1999).

### 1.6. Soluble Phenolic Content

The frozen leaf samples (0.5 g) were rapidly plunged in 20 ml of 80% aqueous ethanol and boiled for 5 min. After filtration through Whatmann no. 1 filter paper, ethanol was eliminated from the filtrate by evaporation in vacuum. Total soluble phenolics in the remaining water phase were determined spectrophotometrically with the Folin-Ciocalteu reagent (prepared by 1:1 dilution with distilled water), against the chlorogenic acid standard (Ferraris et al., 1987).

### 1.7. Superoxide Dismutase Activity

Frozen leaf material (0.5 g) were homogenized in 6 mL 0.1 M potassium phosphate extraction buffer (pH 7, containing 100 mg insoluble PVP and 0.1 mM EDTA) with Ultra Turrax. The homogenate was centrifuged for 5 min at 6000Xg and 4°C. The supernatant was filtered through a Whatman GF/A glass fiber disc with a vacuum filtration system and stored at -70°C (Schöner and Krause, 1990).

SOD activity was determined according to Beyer and Fridovich (1987). The reaction mixture (3 mL) contained potassium phosphate buffer (pH 8, 0.025% Triton X-100 and 0.1 mM EDTA), enzyme extract, 12 mM L-methionine, 75  $\mu$ M nitroblue tetrazolium chloride (NBT)

and 2  $\mu\text{M}$  riboflavin. The reaction mixture was kept under fluorescent light for ten minutes at 25°C. One SOD unit was described as the amount of enzyme where the NBT reduction ratio was 50%. NBT reduction ratios were measured with a spectrophotometer adjusted to 550 nm.

### 1.8. Statistical Processing

All analyses and measurements were made repetitively at least 3 times. The importance levels of the differences between the species belonging to the Lamiaceae and Poaceae families were determined through t-test for each parameter. The averages, standard deviations and t-test results were stated in the tables.

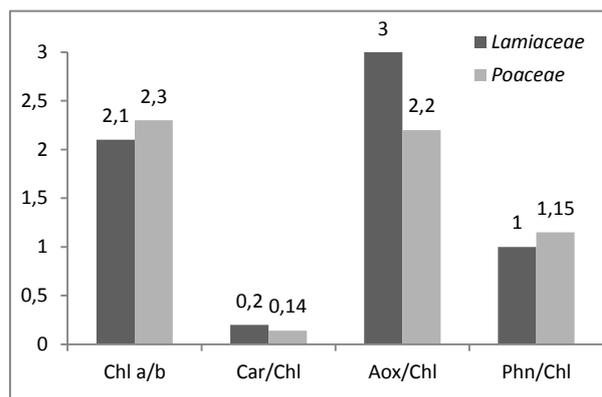
### 3. Results

Average dry weight was determined as 29.5%, average RWC determined as 81.3% at Lamiaceae. The dry weight percentage varies between 15.5% (*Salvia verticillata*) and 46.8% (*Phlomis leucophracta*) at this family (Table 1). The RWC values vary between 55.5% (*Phlomis leucophracta*) and 96.3% (*Salvia verticillata*). Average dry weight was determined as 31.9 %, average relative water content was determined as 74.0 % at Poaceae. The dry weight percentage varies between 15.4 % (*Cynodon* sp.) and 61.3% (*Aegilops speltoides*) at Poaceae. The RWC values vary between 29.3 % (*Aegilops speltoides*) and 97.6 % (*Echinochloa colonum*) at this family. It was observed that the dry weight percentages and the RWC values varied in a wide range in both families. The difference between the families is not statistically significant.

The chlorophyll a, b and total amounts of chlorophyll were analysed at the species belonging to both families. The average amount of chlorophyll a was determined as 1159  $\mu\text{g g}^{-1}$  FW, the average amount of chlorophyll b was determined as 544  $\mu\text{g g}^{-1}$  FW and total average amount of chlorophyll was determined as 1704  $\mu\text{g g}^{-1}$  FW at Lamiaceae. Highest amount of chlorophyll a was identified at *Phlomis leucophracta* species of Lamiaceae, highest amount of chlorophyll b was identified at *Sideritis rubriflora* species of the same family (Table 1). The average amount of chlorophyll a was determined as 1103  $\mu\text{g g}^{-1}$  FW, the average amount of chlorophyll b was determined as 483  $\mu\text{g g}^{-1}$  FW and total average amount of chlorophyll was determined as 1586  $\mu\text{g g}^{-1}$  FW at Poaceae. The greatest amount of chlorophyll a was identified at *Polypogon mospeliensis* species of Poaceae, the highest amount of chlorophyll b was identified at *Phalaris aquatica* species of the same family. The differences identified in respect to the chlorophyll contents and chlorophyll a/b ratio of these two families aren't statistically significant (Table 1, Figure 1).

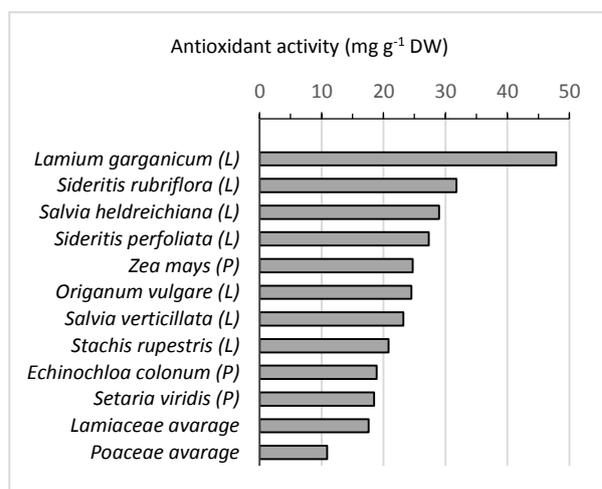
Having separated with thin layer chromatography, the carotenoids were analysed by using  $\beta$ -carotene and their total xanthophyll content was analysed by using spectrophotometer. The average  $\beta$ -carotene was identified as 103  $\mu\text{g g}^{-1}$  FW at Lamiaceae, 84  $\mu\text{g g}^{-1}$  FW at Poaceae. Any statistically significant difference in respect to  $\beta$ -carotene content wasn't determined between these two families. Total average values of the xanthophyll content was measured as 236  $\mu\text{g g}^{-1}$  FW at Lamiaceae, 142  $\mu\text{g g}^{-1}$  FW at Poaceae. While total values of the xanthophyll content vary between 88-382  $\mu\text{g g}^{-1}$  FW at Lamiaceae, vary between 44-304  $\mu\text{g g}^{-1}$  FW at Poaceae. There is a

statistically significant difference ( $P \leq 0.01$ ) between the total values of the xanthophyll content of both families.



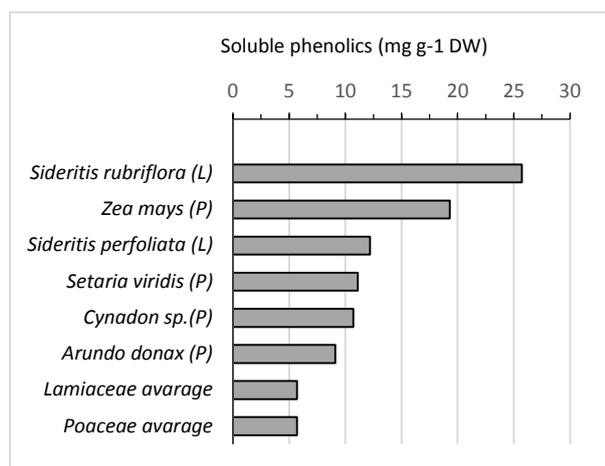
**Fig. 1.** Average values chlorophyll a/b, carotenoids/chlorophyll, total antioxidants/chlorophyll and soluble phenolics/chlorophyll in Lamiaceae and Poaceae families.

Total average antioxidative capacity was measured as 5.19  $\text{mg g}^{-1}$  FW at Lamiaceae, 3,49  $\text{mg g}^{-1}$  FW at Poaceae (Table 2). It was identified that Lamiaceae has higher antioxidant capacity than Poaceae has and this difference is statistically significant at  $P \leq 0.05$  level (Table 2). The species with the highest antioxidative capacity at Poaceae is *Phleum pratense*, the species with the highest antioxidant capacity at Lamiaceae is *Sideritis perfoliata*. Antioxidant/chlorophyll ratio higher at Lamiaceae (Figure 1). The highest antioxidant capacity on the dry weight basis was identified at *Lamium garganicum* species belonging to the Lamiaceae (Figure 2).



**Fig. 2.** Species with high antioxidant activity based on dry weight in Lamiaceae (L) and Poaceae (P) families.

While the phenolic content was identified as 1,67  $\text{mg g}^{-1}$  at Lamiaceae, it was identified as 1,83  $\text{mg g}^{-1}$  at Poaceae. The sample of Lamiaceae family with the highest phenolic content was identified as *Sideritis rubriflora* (5,74  $\text{mg g}^{-1}$ ), the sample of Poaceae family with the highest phenolic content was identified as *Zea mays* (3,82  $\text{mg g}^{-1}$ ) (Table 2). The amount of total phenolic substance is higher at Poaceae as compared with Lamiaceae, however, this difference is not statistically significant. The species with the highest soluble phenolic content on the dry weight basis were identified as *Sideritis rubriflora* and *Zea mays* (Figure 3).



**Fig. 3.** Species with high soluble phenolics based on dry weight in Lamiaceae (L) and Poaceae (P) families,

The average amount of SOD was measured as 225 unit g<sup>-1</sup> FW at Poaceae, as 213 unit g<sup>-1</sup> FW at Lamiaceae. The highest amount of SOD was identified at the *Phlaris pratense* from Poaceae, at the species of *Lamium amplexicaule* from Lamiaceae. Even though SOD content is higher at Poaceae than Lamiaceae, but this difference is not statistically significant.

#### 4. Discussions

The samples belonging to both families were collected in similar ecological conditions and in the same vegetative period. The average dry weight values of species of Lamiaceae and Poaceae families were identified as approximate to each other. However, significant differences were determined in respect to dry weight values among the members of the same family. These differences can be explained with soil, water conditions and microclimate differences (Oncel et al., 2004). That relative water content is higher at Lamiaceae's species can be associated with better water accession and better water conservation. It was reported in the study intending to determine the relationship between soil saltiness and RWC in the species of 6 turf (Poaceae) that the more soil saltiness increases, reduces RWC rates (Uddin et al., 2012).

Though the chlorophyll content significantly changes among the members of the same family, the difference between Lamiaceae and Poaceae is not significant. However, that the values of chlorophyll b is higher at Lamiaceae leads to the ratio of a/b to be increased in favor of Poaceae (Figure 1). Castrillo et al. (2001) investigated the contents of chlorophyll of 11 species belonging to Lamiaceae. They identified the ratios of a/b above 2.0 at two species belonging to Nepetoideae subfamily, below 2.0 (1.41-1.76) at other species as well. The contents of chlorophyll of 11 species belonging to Poaceae were analyzed and it was identified that the ratios of chlorophyll a/b varied between 3.5 and 2.0 (Uddin et al., 2012). The chlorophyll content and ratio of chlorophyll a/b is associated with receive sunlight condition of plants. The lower ratio of chlorophyll a/b is expected in plants growing in the shade.

According to the findings of this study, the amounts of both  $\beta$ -carotene and xanthophyll of Lamiaceae are higher than those of Poaceae (Table 2). The ratio of carotenoids

per chlorophyll was found as 0.2 at Lamiaceae 0.14 at Poaceae (Figure 1). Carotenoids can be asserted to have a more important role in protection of photosynthetic system at the members of Lamiaceae. These findings are more different than those of Oncel et al. (2004) who identified low-carotenoid contents at the species of Lamiaceae growing at steppe. Rather low amount of carotenoids (0.34  $\mu\text{g g}^{-1}$  DW  $\beta$ -carotene and 1.3  $\mu\text{g g}^{-1}$  DW xanthophyll) was identified through HPLC analysis in the edible parts of the *Thymus vulgaris* plant from Lamiaceae. However, it was identified in the same study that the reference values of this species related carotenoids were 29  $\mu\text{g g}^{-1}$   $\beta$ -carotene and 19  $\mu\text{g g}^{-1}$  xanthophyll (El-Qudah, 2014). In a comparative study, the contents of carotenoid were identified as 260-430  $\mu\text{g g}^{-1}$  FW at 10 species of Poaceae family growing at tundra (Markowskaya et al., 2012). These high values of carotenoid can be resulted in due to tundra conditions.

The antioxidative capacity of the species of Lamiaceae investigated in this study was identified higher than that of the species of Poaceae (Table 2). The antioxidant capacity per chlorophyll is significantly high at Lamiaceae (Figure 1). The total antioxidative capacity on the dry weight basis is significantly high at *Lamium garganicum* species (Figure 2). Matkowski et al. (2008) identified a high antioxidative activity at the leaves of three species of *Salvia* (*S. mitiorrhiza*, *S. przewalskii* and *S. verticillata*). The authors offered the species of *Salvia* as source of potential antioxidant. But it was determined in the current study that the species of *Sideritis* have greater antioxidative capacity than the species of *Salvia* have. It was reported that 4 species of Lamiaceae growing naturally in Turkey, have significantly high antioxidative capacity and this situation is associated especially with content of polyphenol (Erdemoglu et al. 2006). The high values were identified in the study in which, antioxidative activities of the species of plant being known as *Salvia* and used as a drink in Turkey such that gallic acid equivalent was 130 mg g<sup>-1</sup> at *Salvia fruticosa*; 154 mg g<sup>-1</sup> at *Sideritis congesta*, 120 mg g<sup>-1</sup> at *Sideritis pisidica* (Erdoğan et al., 2010).

While total soluble phenolic contents of the species of Lamiaceae family distribute in a wide range, the phenolic values of the species of Poaceae were identified as approximate to each other (Table 2). A small difference in favor of Poaceae was identified with regard to soluble phenolic compounds per chlorophyll (Figure 1). It was revealed in a study which subject was antioxidative and antimicrobial activities of the species of *Echinochloa colona* belonging to Poaceae that this species showed high level of antioxidant activity. Total phenolic content of this species was stated as 734 mg ml<sup>-1</sup> (Ajaib et al., 2013). It was stated that the antioxidative activity of corn silk (*Zea sp.*) was resulted from high contents of phenol and flavonoid and its total phenolic content was identified as 2.78 mg g<sup>-1</sup> (Ebrahimzadehet et al., 2008). The species of poaceae family with high total contents of soluble phenolic were identified as *Zea mays* and *Arundo donax* (respectively 3.82 and 3.01 mg g<sup>-1</sup> FW) in our study. The content of phenolic on the dry weight basis was identified significantly high at the species of *Zea mays*. These values are approximate to those identified by Ebrahimzadeh et al. 2008.

It was reported in the studies investigating antioxidant activity of the species of *Salvia* growing in Turkey that the species with high total content of phenol have high antioxidant activity (Albayrak et al., 2008; Erdemoglu et al., 2006). Especially high values of the phenolic

substances such as carsonic acid, rosmarinic acid, salvianolic acid are responsible for the antioxidative activity at the species of *Salvia* (Lu and Foo, 2002; Triantaphyllou et al., 2001).

**Table 1.** List of species from Lamiaceae and Poaceae, it's localities and their dry weight, RWC and chlorophyll values (*P* values were not shown any significant difference).

| <i>Lamiaceae species and locality</i>                       | Dry Weight (%)   | RWC (%)          | Chl-a ( $\mu\text{g g}^{-1}$ FW) | Chl-b ( $\mu\text{g g}^{-1}$ FW) | Total Chl. ( $\mu\text{g g}^{-1}$ FW) |
|---|------------------|------------------|----------------------------------|----------------------------------|---------------------------------------|
| <i>Calamintha nepeta</i> (Apsun, Gözne)                     | 24.6±3.3         | 74.3±6.1         | 1000±35                          | 516±30                           | 1517                                  |
| <i>Lamium amplexicaule</i> (Bükdeğirmeni)                   | 19.1±1.8         | 95.7±3.9         | 776±48                           | 379±22                           | 1155                                  |
| <i>Lamium garganicum</i> (Değirmençay)                      | 16.7±3.0         | 88.0±5.2         | 1402±41                          | 646±27                           | 2048                                  |
| <i>Marrubium vulgare</i> (Aydıncık, Bükdeğirmeni, Apsun)    | 27.4±2.2         | 64.8±2.9         | 1290±57                          | 585±35                           | 1876                                  |
| <i>Micromeria myrtifolia</i> (Kuyuluk, Değirmençay)         | 33.9±0.7         | 85.1±3.4         | 990±18                           | 493±27                           | 1484                                  |
| <i>Origanum vulgare</i> (Kuyuluk, Bükdeğirmeni)             | 29.0±1.3         | 85.4±3.2         | 701±64                           | 365±38                           | 1080                                  |
| <i>Phlomis leucophracta</i> (Bükdeğirmeni)                  | 46.8±2.5         | 55.5±3.8         | 2055±67                          | 866±41                           | 2921                                  |
| <i>Salvia frigida</i> (Çamlıyayla- Çuvalgı)                 | 35.0±2.8         | 89.4±4.1         | 1285±30                          | 591±24                           | 1876                                  |
| <i>Salvia heldreichiana</i> (Apsun)                         | 23.8±1.3         | 89.3±1.7         | 579±36                           | 307±22                           | 885                                   |
| <i>Salvia verticillata</i> (Kuyuluk)                        | 15.5±1.4         | 96.3±3.2         | 941±27                           | 471±24                           | 1413                                  |
| <i>Salvia virgata</i> (Apsun)                               | 17.8±1.0         | 83.2±3.1         | 882±31                           | 460±26                           | 1342                                  |
| <i>Sideritis perfoliata</i> (Kuyuluk)                       | 43.3±0.5         | 93.5±3.7         | 1774±45                          | 560±31                           | 2335                                  |
| <i>Sideritis rubriflora</i> (Işıklı)                        | 22.3±0.5         | 84.8±3.5         | 1952±34                          | 924±15                           | 2876                                  |
| <i>Stachys rupestris</i> (Apsun)                            | 38.9±1.9         | 62.9±4.4         | 1243±31                          | 591±47                           | 1834                                  |
| <i>Teucrium chamaedrys</i> (Çamlıyayla- G.)                 | 32.0±0.9         | 85.6±1.9         | 906±27                           | 524±38                           | 1430                                  |
| <i>Teucrium polium</i> (Aydıncık, Bükdeğirmeni, Çamlıyayla) | 45.8±3.4         | 67.4±3.0         | 762±54                           | 428±37                           | 1190                                  |
| <b>Lamiaceae Avarage</b>                                    | <b>29.5±10.1</b> | <b>81.3±12.1</b> | <b>1159±443</b>                  | <b>544±160</b>                   | <b>1704±583</b>                       |
| <b>Poaceae species and locality</b>                         |                  |                  |                                  |                                  |                                       |
| <i>Aegilops speltoides</i> (Çamlıyayla)                     | 61.3±0.8         | 29.3±0.9         | 555±48                           | 319±27                           | 874                                   |
| <i>Arundo donax</i> (Kazanlı)                               | 32.9±1.3         | 87.9±2.9         | 584±115                          | 261±73                           | 845                                   |
| <i>Avena sterilis</i> (Çamlıyayla)                          | 42.8±3.0         | 49.2±1.5         | 617±122                          | 308±80                           | 925                                   |
| <i>Brachypodium pinnatum</i> (Çamlıyayla- G.)               | 40.9±2.7         | 78.2±2.5         | 1316±44                          | 574±36                           | 1890                                  |
| <i>Brachypodium sylvaticum</i> (Çamlıyayla)                 | 39.9±1.9         | 79.0±1.1         | 606±80                           | 336±86                           | 942                                   |
| <i>Cynodon dactylon</i> (Kazanlı)                           | 19.3±2.6         | 84.7±2.2         | 1443±54                          | 637±41                           | 2080                                  |
| <i>Cynodon sp.</i> (Kazanlı)                                | 15.4±1.1         | 94.2±0.7         | 1006±45                          | 434±52                           | 1440                                  |
| <i>Echinochloa colonum</i> (Kazanlı)                        | 19.0±0.6         | 97.6±3.3         | 1078±68                          | 489±43                           | 1567                                  |
| <i>Echinochloa crusgalli</i> (Kazanlı)                      | 19.0±0.9         | 93.2±3.2         | 1045±45                          | 437±37                           | 1482                                  |
| <i>Phalaris aquatica</i> (Kazanlı)                          | 37.2±3.0         | 56.3±2.0         | 1456±56                          | 727±41                           | 2183                                  |
| <i>Phleum pratense</i> (Çamlıyayla)                         | 54.6±4.2         | 44.6±2.9         | 1006±77                          | 481±45                           | 1487                                  |
| <i>Polypogon monspeliensis</i> (Çamlıyayla)                 | 27.2±2.5         | 77.6±3.7         | 1893±82                          | 713±76                           | 2606                                  |
| <i>Setaria viridis</i> (Yenişehir)                          | 17.8±1.5         | 80.6±5.5         | 1670±57                          | 617±52                           | 2287                                  |
| <i>Zea mays</i> (Kazanlı)                                   | 19.8±1.7         | 93.2±3.2         | 1169±48                          | 429±35                           | 1598                                  |
| <b>Poaceae Avarage</b>                                      | <b>31.9±14.2</b> | <b>74.0±20.0</b> | <b>1103±406</b>                  | <b>483±146</b>                   | <b>1586±546</b>                       |
| <b>Statistics (T-test, P values)</b>                        | <b>0.60</b>      | <b>0.24</b>      | <b>0.73</b>                      | <b>0.30</b>                      | <b>0.58</b>                           |

**Table 2.** Antioxidant compounds content and SOD activities of species from Lamiaceae and Poaceae families (\*significant at  $P \leq 0.05$  and \*\* significant  $P \leq 0.01$ ).

| <i>Lamiaceae</i>  | $\beta$ -Caroten<br>( $\mu\text{g g}^{-1}$ FW) | Xanthophyll<br>( $\mu\text{g g}^{-1}$ FW) | Antioxidant<br>capacity<br>( $\text{mg g}^{-1}$ FW) | Soluble<br>phenolics<br>( $\text{mg g}^{-1}$ FW) | Total SOD<br>(Unit $\text{g}^{-1}$ FW) |
|---|--|---|---|--|--|
| <i>Calamintha nepeta</i> (Apsun, Gözne)                     | 82±6.4   | 290±21.0                                  | 4.1±0.43  | 1,26 ± 0,07                                      | 197±32                                 |
| <i>Lamium amplexicaule</i> (Bükdeğirmeni)                   | 88±8.7   | 186±12.3                                  | 2.6±0.41  | 1,07 ± 0,08                                      | 296±20                                 |
| <i>Lamium garganicum</i> (Değirmençay)                      | 164±14.4                                       | 336±16.8                                  | 8.0±0.22  | 0,75 ± 0,05                                      | 186±26                                 |
| <i>Marrubium vulgare</i> (Aydıncık, Bükdeğirmeni, Apsun)    | 114±15.2                                       | 265±24.0                                  | 4.1±0.71  | 1,26 ± 0,05                                      | 193±45                                 |
| <i>Micromeria myrtifolia</i> (Kuyuluk, Değirmençay)         | 160±15.0                                       | 232±18.1                                  | 2.8±0.19  | 1,27 ± 0,05                                      | 193±23                                 |
| <i>Origanum vulgare</i> (Kuyuluk, Bükdeğirmeni)             | 89±11.0  | 124±19.2                                  | 7.1±0.27  | 0,98 ± 0,05                                      | 185±45                                 |
| <i>Phlomis leucophracta</i> (Bükdeğirmeni)                  | 116±22.1                                       | 368±19.8                                  | 3.0±0.25  | 3,2 ± 0,07                                       | 175±14                                 |
| <i>Salvia frigida</i> (Çamlıyayla- Çuvalı)                  | 124±11.4                                       | 294±32.8                                  | 3.6±0.91  | 0,55 ± 0,07                                      | 181±16                                 |
| <i>Salvia heldreichiana</i> (Apsun)                         | 86±6.6   | 165±13.5                                  | 6.9±0.33  | 1,45 ± 0,01                                      | 235±45                                 |
| <i>Salvia verticillata</i> (Kuyuluk)                        | 136±13.6                                       | 382±22.5                                  | 3.6±0.54  | 0,39 ± 0,08                                      | 227±30                                 |
| <i>Salvia virgata</i> (Apsun)                               | 66±8.0   | 187±23.9                                  | 2.2±0.57  | 0,14 ± 0,09                                      | 266±37                                 |
| <i>Sideritis perfoliata</i> (Kuyuluk)                       | 148±12.5                                       | 340±26.0                                  | 11.8±0.59   | 5,3 ± 0,07                                       | 188±35                                 |
| <i>Sideritis rubriflora</i> (Işıklı)                        | 92±8.4   | 304±22.4                                  | 7.1±0.25  | 5,74 ± 0,07                                      | 221±40                                 |
| <i>Stachys rupestris</i> (Apsun)                            | 44±8.5   | 92±15.2                                   | 8.1±0.50  | 0,9 ± 0,08                                       | 204±22                                 |
| <i>Teucrium chamaedrys</i> (Çamlıyayla- G.)                 | 82±7.6   | 157±13.5                                  | 2.3±0.08  | 0,23 ± 0,1                                       | 258±40                                 |
| <i>Teucrium polium</i> (Aydıncık, Bükdeğirmeni, Çamlıyayla) | 63±9.2   | 88±7.5                                    | 5.4±0.24  | 2,17 ± 0,08                                      | 197±24                                 |
| <b>Lamiaceae Avarage</b>                                    | <b>103±35</b>                                  | <b>236±95</b>                             | <b>5.19±2.62</b>                                    | <b>1.67±1.63</b>                                 | <b>213±34</b>                          |
| <b>Poaceae</b>  |  |   |   |  |  |
| <i>Aegilops speltoides</i> (Çamlıyayla)                     | 92±12.7  | 127±7.1                                   | 4.8±0.07  | 1,7 ± 0,05                                       | 209±14                                 |
| <i>Arundo donax</i> (Kazanlı)                               | 24±11.3  | 72±6.9                                    | 3.4±0.58  | 3,01 ± 0,14                                      | 234±74                                 |
| <i>Avena sterilis</i> (Çamlıyayla)                          | 68±7.6   | 84±8.0                                    | 4.1±0.27  | 1,94 ± 0,08                                      | 229±28                                 |
| <i>Brachypodium pinnatum</i> (Çamlıyayla- G.)               | 84±15.9  | 52±7.1                                    | 1.7±0.30  | 1,48 ± 0,05                                      | 188±62                                 |
| <i>Brachypodium sylvaticum</i> (Çamlıyayla)                 | 36±6.4   | 115±9.5                                   | 4.8±0.99  | 1,58 ± 0,25                                      | 214±54                                 |
| <i>Cynodon dactylon</i> (Kazanlı)                           | 128±18.6                                       | 232±8.0                                   | 2.9±0.57  | 1,35 ± 0,01                                      | 223±39                                 |
| <i>Cynodon sp.</i> (Kazanlı)                                | 88±8.1   | 132±10.2                                  | 2.2±0.40  | 1,65 ± 0,33                                      | 161±24                                 |
| <i>Echinochloa colonum</i> (Kazanlı)                        | 132±16.7                                       | 146±4.7                                   | 3.6±0.44  | 1,43 ± 0,13                                      | 210±22                                 |
| <i>Echinochloa crusgalli</i> (Kazanlı)                      | 60±12.1  | 97±5.2                                    | 2.2±0.46  | 1,43 ± 0,06                                      | 206±60                                 |
| <i>Phalaris aquatica</i> (Kazanlı)                          | 88±10.3  | 304±26.8                                  | 1.9±0.18  | 1,71 ± 0,07                                      | 295±62                                 |
| <i>Phleum pratense</i> (Çamlıyayla)                         | 52±10.8  | 148±10.4                                  | 5.8±0.43  | 1,33 ± 0,08                                      | 296±49                                 |
| <i>Polypogon monspeliensis</i> (Çamlıyayla)                 | 104±7.9  | 294±11.8                                  | 3.4±0.44  | 1,16 ± 0,15                                      | 196±34                                 |
| <i>Setaria viridis</i> (Yenişehir)                          | 180±7.9  | 158±15.1                                  | 3.3±0.43  | 1,98 ± 0,04                                      | 241±28                                 |
| <i>Zea mays</i> (Kazanlı)                                   | 55±17.1  | 44±7.7                                    | 4.9±0.37  | 3,82 ± 0,14                                      | 253±24                                 |
| <b>Poaceae Avarage</b>                                      | <b>84±42</b>                                   | <b>142±84</b>                             | <b>3.49±1.22</b>                                    | <b>1.83±0.70</b>                                 | <b>225±36</b>                          |
| <b>Statistics (T-test, P values)</b>                        | <b>0.23</b>                                    | <b>0.006**</b>                            | <b>0.04*</b>  | <b>0.74</b>                                      | <b>0.34</b>                            |

The amounts of total phenolic were determined as *Micromeria croatia* 13,6 %, *M. juliana* 10,8%, *M. thymifolia* 9,7% in the analyse performed with dried plant samples at the species of Lamiaceae growing in Croatia. It was stated that there was a strong positive correlation between phenolic compounds and antioxidative activity (Knezevic et al. 2011). High content of substance such as  $13.4 \text{ mg g}^{-1}$  FW was identified at *Lavandula angustifolia*, in a study in which total content of phenolic of four species of Lamiaceae were investigated (Swedan, 2013). The reason why total content of phenolic of the species of *Salvia* have been identified as high in the literature is that extraction was performed with acid hydrolysis. Since acid hydrolysis wasn't used in our study, phenolic values with low solubility at the species of *Salvia*, high solubility at

the species of *Sideritis* were identified. The highest values of soluble phenolic at dry weight basis were identified at *Sideritis rubriflora* (Figure 3). The average total amount of soluble phenolic is higher at Poacea than Lamiaceae. But the highest values of soluble phenolic were identified at two species of *Sideritis*. The high values of phenolic existing at some species of Lamiaceae may lead these species to be used as food, syrup and tea. The reasons why the species of Lamiaceae have been intensely investigated are phenolic compounds cause the inhibition of the production of fungal enzyme and of the enzymes produced by pathogens and removal of free radicals.

Unsuitable environmental conditions cause oxidative stress in plants. SOD enzyme has a critical role in

protection of plants against oxidative stress (Keleş and Everest, 2008). Cross tolerance occurs in the plants being affected simultaneously from a great number of stress factors in natural conditions. One of the important components of cross tolerance is an increase in SOD activity (Bowler et al., 1992). In the study in which effects of both temperature and water stress at two species of wheat were investigated, the highest SOD activities were identified when temperature and water stress affect together (Keleş and Öncel, 2000). In the study in which the plants adapted to high mountains and steppe conditions were compared in regard to their SOD activities; while the average of SOD activity was determined as 213 units in high mountain plants, it was determined as 134 units in steppe plants (Öncel et al., 2004). While SOD activity was identified as high at the species of Poaceae invested in the same study, it was identified as quite changeable at the species of Lamiaceae. The SOD activity was identified as 225 units at the species

of Poaceae and 213 units at the species of Lamiaceae in the current study. The findings indicate that The SOD activity has been affected from habitat features of plants rather than their genetic features.

The antioxidants showing directly or indirectly radical scavenging effect, highly exist at the species of Poaceae that can be made use of feed plant, at the Sideritis species of Lamiaceae that can be made use of drink as well.

Moreover, although the species of Stachys hasn't widely used and investigated, it is among the species containing the highest antioxidant activity. The members of Lamiaceae, which are valuable in aromatic compounds and contents of volatile oil, have been intensively investigated by the science environment. It was concluded that the species of Poaceae are especially rich in soluble phenolic compounds and they should be further investigated.

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