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Research article/Araştırma makalesi

Changes in morphology and physiology of *Convolvulus oleifolius* var. *Deserti* in different habitats

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Abstract

Morphological features, photosynthetic pigment contents, proline and total protein amounts of *Convolvulus oleifolius* Desr. var. *deserti* Pamp./Surmeli yayilgan in different vegetation zones of Apostol Island are studied. Samples were collected from littoral-epilittoral and interior vegetation zones of Apostol Island, Bodrum-Turkbuku-Mugla-Turkey. There are obvious differences in terms of stem length, organ size, hair density and inflorescence. Photosynthetic pigments in littoral-epilittoral vegetation zone are less than in interior vegetation zone. Prolin amount is significantly higher, but total protein amount is significantly lower in littoral-epilittoral vegetation zone than in interior vegetation zone. It may be thought that the morphological and physiological features of the *C. oleifolius* var. *deserti* exhibited considerable differences which appeared be the product of adaptive mechanisms to different habitat. So, our study is important for improving and selecting crops tolerant of and/or adapted to different habitat.

Key words: : *Convolvulus*, Photosynthetic pigment, proline, protein, vegetation zone

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Farklı habitatlarda *Convolvulus oleifolius* var. *Deserti*'nin morfoloji ve fizyolojisinde değişimler

Özet

Apostol adasının farklı vejetasyon zonlarında bulunan *Convolvulus oleifolius* Desr. var. *deserti* Pamp./Sürmeli yayılgan'ın morfolojik özellikleri, fotosentetik pigment içerikleri, prolin ve toplam protein miktarları çalışılmıştır. Örnekler Bodrum-Türkbükü-Muğla-Türkiye, Apostol adasının littoral-epilittoral ve interior vejetasyon zonlarından toplanmıştır. Gövde uzunluğu, organ büyüklüğü, tüy yoğunluğu ve çiçeklenme durumunda belirgin farklılıklar vardır. Fotosentetik pigmentler littoral-epilittoral vejetasyon zonda, interior vejetasyon zondakinden daha azdır. Littoral-epilittoral vejetasyon zonda prolin miktarı interior vejetasyon zondakinden önemli derecede daha yüksekken, toplam protein miktarı önemli derecede daha düşüktür. *C. oleifolius* var. *deserti*'nin morfolojik ve fizyolojik özelliklerinin çeşitli habitatlara uyum mekanizması sonucu ortaya çıkan önemli farklılıkları sergilediği düşünülebilir. Bu nedenle, çalışmamız farklı habitatlara uyumlu ve/veya toleranslı bitkilerin seçilmesi ve değerlendirilmesi için önemlidir.

Anahtar kelimeler: *Convolvulus*, Fotosentetik pigment, Prolin, Protein, Vejetasyon zonu

1. Introduction

The family Convolvulaceae (bindweed or morning glory) are a group of about 60 genera and more than 1.650 species of mostly herbaceous climbers, herbs, shrubs and rarely trees, some species with milky sap (Austin and Zosimo, 1996). It is presented throughout temperate and tropical regions of the world, and has a wide range of habitats (Heywood, 1985). According to Flora of Turkey, belong to 33 species (36 taxa) of genus *Convolvulus* were distributed in Turkey (Paris, 1978; Davis et al., 1988).

Convolvulus oleifolius Desr. var. *deserti* Pamp./Surmeli yayilgan (Güner et al., 2012) is a woody based prennials, shrublets or shrubs and distributing rocky and stony slopes, macchie areas, phrygana and sand dunes near the sea in Agean and Mediterranean regions in Turkey (Aykurt and Sümbül, 2010).

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Excessive water, water temperature and especially salinity are environmental stress for plants in coastal areas. In those environments, seawater infiltrations can occur or the sea provides the only source of water for irrigation (Baccio et al., 2004). The injurious effects of salinity are associated with water deficit, ionic imbalance, stomatal behaviour, photosynthetic efficiency, protein reduction, proline accumulation and oxidative damage (Parida et al., 2002; Ashraf and Haris, 2004). This kind of response leads to ecotype differentiation (Amzallag et al., 1993). Understanding the physiological bases of salinity adaptation could help us to improve and select crops tolerant of and/or adapted to different habitat.

In the present study, some morphological and physiological differences between *C. oleifolius* var. *deserti* in littoral-epilittoral and interior vegetation zones of Apostol Island in Bodrum-Turkbuku-Mugla are presented.

2. Materials and methods

2.1. Sample collection

Convolvulus oleifolius Desr. var. *deserti* Pamp. samples were colllected during spring and summer from littoral-epilittoral and interior vegetation zones in Apostol Island in Bodrum-Turkbuku-Mugla-Turkey for determination of morphological and physiological differences by author (Senol, 2006). All herbarium samples deposited in Herbarium EGE.

2.2. Morphological characters

All morphological data presented and used in the description were directly observed by the authors. All the morphological characters traditionally used to identify. Convolvulus species were studied and compared with other members of sect. Convolvulus. Features of gross morphology were exemined under a stereoscopic microscope. Also, growth parameters of ten plants were measured for each vegetation zone.

2.3. Photosynthetic pigment content

Chlorophyll and carotenoid concentration were determined from fresh leaves. A fresh leaf sample of 0.1 g was ground and extracted with 5 ml of 80% (v/v) acetone in the dark (Arnon, 1949). The mixture was filtered and absorbancies (Jenway 6105 UV/VIS, Spectrophotometer) were determined at 645, 663 and 450 nm. Concentration of chlorophyll a (Chl a), chlorophyll b (Chl b) and carotenoids were estimated by the equations of Witham et al. (1971).

2.4. Proline content

Proline content was determined according to the modified method of Bates et al. (1973). Fresh leaf samples were homogenized in 3% (w/v) sulfosalicylic acid solution and then centrifuged. The supernatant was taken into a test tube to which glacial acetic acid and acid ninhydrin solution were added. Tubes were incubated in a boiling water bath for 1 h and then allowed to cool to room temperature. After adding toluene, the mixture was vortexed and allowed to separation of toluene and aqueous phase. The absorbance of toluene phase was measured at 520 nm in a spectrophotometer. The concentration was calculated from a proline standard curve and expressed as μmol/g FW.

2.5.Total protein

Total soluble protein was extracted from fresh leaves and quantified by the Bradford protein assay (1976) using BSA as a standard. In the Bradford assay, protein concentration is determined by quantifying the binding of the dye, Coomassie Brilliant Blue G-250, to the unknown protein solution, as compared to known standards. Tubes containing 100 μl aliquots of known concentrations of Bovine Serum Albumin (BSA; 0.156 mg l-1 to 10 mg l-1 in 0.15 M NaCl), were prepared. Blank tubes containing 100 μl of 0.15 M NaCl were also prepared. One ml Coomassie Brilliant Blue solution was added to each tube and the mixtures vortexed. The reactions were left at room temperature for 2 min. The absorbance at wavelength of 595 nm was determined against the blank and the standard curve of absorbance versus protein concentration plotted (Copeland, 1994).

2.6. Statistical analysis

All the results were statistically analyzed through independent-samples T test at $p<0.05$ level.

3. Results

During the study of Flora and Vegetation in South Aegean Sea Islands (Çeşme-Antalya), *C. oleifolius* var. *deserti* were collected from Mugla-Turkbuku Apostol Island (Senol, 2006) (Figure 1). Three vegetation zones (littoral, epilittoral and interior) were distinguished and described in the island. The littoral vegetation zone was restricted to calcerous rocky up to 3-4m high above sea level and was characterized by a loose floristic composition. Other plant taxa in this zone accompanying study materials were *Lotus cytisoides* L., *Limonium gmelini* (Willd.) Kuntze, *Frankenia hirsuta* L., *Hilimione portulacoides* (L.) Allen, *Malcolmia flexuosa* (Sibth. Et Sm.) Sibth. Et Sm.. Also, this zone was exposed to the wave effect. Epilittoral vegetation zone was only weakly influenced by wave action and sea spray. Closeness of vegetation in this zone is 50-60% and accompanying other taxa were *Helichrysum stoechas* (L.) Moench subsp. *barrelieri* (Ten.) Nyman. and *Phagnalon graecum* Boiss.. After a height of 8-10m, the calcareous interior vegetation zone took place. In this zone, 3-4 m tall shurb communities dominated by *Olea europeae* L. and *Juniperus phoenica* L. were observed and other woody species were *Pistacia lentiscus* L., *Spartium junceum* L., *Phillyrea latifolia* L. (Senol, 2006).

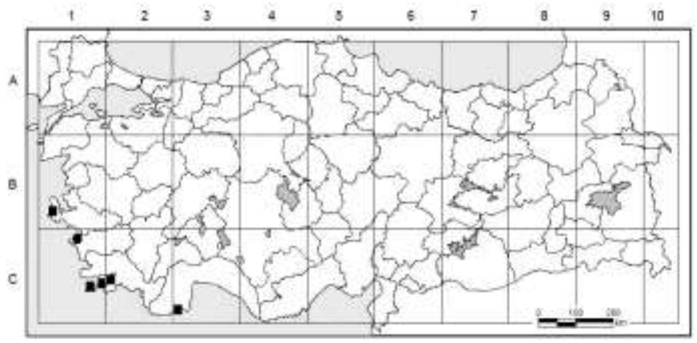

Figure 1. (■) Distribution of *C. oleifolius* var. *deserti* according to Aykurt and Sümbül (2010), (●) Apostol Islands where the study species were collect

C. oleifolius var. *deserti* samples collected from littoral-epilittoral vegetation zone were 15-20 cm, woody-based shrublets and scattered branched. It had a dense white indumentum. Inflorescence was axillary and solitary (Figure 2). The samples collected from interior vegetation zone were 40-50cm, woody-based shrub. It had sparsely silvery pilose. Infloresence was axillary and terminal, especially 4-5 flowered cymes (Figure 3).

Figure 2. Habitat of *C. oleifolius*. var. *deserti* in littoral-epilittoral vegetation zone

Figure 3. Habitat of *C. oleifolius* var. *deserti* in interior vegetation zone

Each of the three vegetation zones are on calcareous rocks. The plants were located in the rock cracks in littoral-epilittoral zone, but on calcareous soils formed from the host rock in interior zone. Samples were collected from the same slope and the same direction of Apostol Island. The temperature difference was very small. The most important factor of habitat differences was the distance from the sea. Littoral-epilittoral vegetation zone exposed to sea water spray. Members of the species to cope with salt spray, have developed adaptation mechanisms and demostrated some morphological and physiological differences.

Table 1. summarize the differences between littoral-epilittoral and interior vegetation zones in morphology of *Convolvulus*. The results clearly show that seven statistically significant morphological differences are recorded. Also, plant forms, inflorescence and hair density are different.

Table 1. Morphological differences between *Convolvulus oleifolius* var. *Derserti* in littoral-epilittoral and interior vegetation zones (n=10)

	Littoral-Epilittoral Zone	Interior Zone
Shoot Fresh Weight (g)	22.2540±8.0914	61.9640±16.953*
Shoot Dry Weight (g)	2.0549±1.1413	7.6717±2.0188*
Root Fresh Weight (g)	4.0320±0.8464	12.9000±1.0040*
Root Dry Weight (g)	1.3066±0.0750	1.4275±0.1120
Leaf Fresh Weight (g)	0.0130±0.0016	0.0607±0.0157*
Leaf Dry Weight (g)	0.0024±0.0003	0.0118±0.0010*
Leaf number	39.2000±3.5637	42.4000±0.5477
Leaf area (mm)	30.0610±1.7006	181.8576±35.021*
Stem length (cm)	15-20	40-50*
Plant form	Woody based shrublest	Woody based shrub
Inflorescence	Axillary-solitary	Axillary -terminal
Hairs	Dense white indimentum	Sparsely silvery pilose

Within each line, * Significantly different at alpha 0.05 according to independent-samples T test.

Table 2. Physiological differences between *Convolvulus oleifolius* var. *deserti* in littoral-epilittoral and interior vegetation zones (n=4)

	Littoral-Epilittoral Zone	Interior Zone
Chl a (mg ml^{-1})	0.3859±0.1353	0.5640±0.2072
Chl b (mg ml^{-1})	0.3966±0.1061	0.7422±0.3333
Total Chl (mg ml^{-1})	0.7822±0.2331	1.3058±0.5371
Carotenoid (mg ml^{-1})	3.3533±0.8068	5.0356±1.6139
Prolin (µmol g^{-1})	8.8329±0.7016	5.5392±1.3700*
Total Protein (mg ml^{-1})	0.9880±0.1449	4.0484±0.9144*

Within each line, * Significantly different at alpha 0.05 according to independent-samples T test.

Tolerance to environmental stresses as salinity of plants can be determined by using different parameters. Chlorophyll a, b and carotenoids in the fresh leaves of the *C. oleifolius* var. *deserti* are presented in Table 2. The results show that, chlorophyll a- b, total chlorophyll and carotenoids in littoral-epilittoral vegetation zone are less than in interior vegetation zone. However the differences are not significant . Some studies have shown that leaf chlorophyll and carotenoid contents were reduced in salt stress (Sultana et al., 2002; Sabir et al., 2009; Turkyilmaz, 2012). The observed chlorophyll depletion may be considered to be a result of the inhibition of chlorophyll biosynthesis (Khan, 2003).

Prolin amount is significantly higher, but total protein amount is significantly lower in littoral-epilittoral vegetation zone than in interior vegetation zone (Table 2). Aghaleh et al. (2009) showed that salinity triggered some solutes (proline and soluble saccharides) accumulation and blocked protein production in *Salicornia persica* and *S. europaea*. High level of proline content can be considered beneficial to stressed plants. Significant correlation between enhanced tolerance and proline accumulation in plants under saline condition has been reported in Ashraf and Foolad's study (2007). Plants need to have special mechanisms for adjusting internal osmotic conditions and changing of osmotic pressure in the root environment. In salt stress plants osmotic potential of vacuole decreased by proline accumulation (Yoshiba et al., 1997). The protein content showed a statistically significant decrease upon salt treatment in *Aegiceras corniculatum* (Parida et al., 2005) too. Agastian et al. (2000) reported that proteins level decreased under salinity is due to low uptake of nitrate ions.

It is worth mentioning that the morphological and physiological features of the recognized plants exhibited considerable differences which appeared be the product of the adaptive mechanisms to environment (Batanouny et al., 1991; Morsy, 2008).

4. Conclusions

There are obvious differences in terms of stem length, organ size, hair density, prolin and total protein amounts between *C. oleifolius* Desr. var. *deserti* Pamp. in littoral-epilittoral and interior vegetation zones. Other species like *Limonium gimelinii*, *Frankenia hirsuta*, *Malcomia flexiosa* and *Lotus cytisoides* that share the littoral vegetation zone in the study area, especially in terms of size is likely to see the differences. So that the perennial sea water-resistant individuals are exposed to salt stress, the reaction can be expressed by. These differences can be the product of genetic responses of their habitats. The results have demonstrated that morphological and physiological differences can play an important role in the adjustment of this genus to various habitats. Also, knowing these differences provide us to improve and select crops tolerant of and/or adapted to different habitat.

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