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Annals of Biological Research, 2011, 2 (5) :164-171  
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ISSN 0976-1233  
CODEN (USA): ABRNBW

# Effects of UV-A and UV-C radiation on some morphological and physiological parameters in Savory (*Satureja hortensis L.*)

Parisa Rahimzadeh<sup>1\*</sup>, Siavash Hosseini<sup>2</sup>, Kamalaadin Dilmaghani<sup>1</sup>

<sup>1</sup>Department of Biology, Islamic Azad University, Marand Branch, Marand, Iran

<sup>2</sup>Department of Biology, Urmia University, Urmia, Iran

## ABSTRACT

The present study was conducted to assess the effects of UV-A and UV-C on some morphological and physiological parameters of savory (*Satureja hortensis L.*). Plants were grown in a uniform environment and after 50 days they were exposed to UV-A and UV-C radiation for 15 and 8 days respectively. Alterations in growth parameters, photosynthetic pigments, UV absorbing compounds and protein and carbohydrate contents were measured. UV-C-treated plants showed a significant decrease in shoot growth, leaf number and shoot fresh and dry weights as well as leaf protein, leaf carbohydrate, chlorophyll *a*, *b* and total and carotenoid contents. Also treatment with UV-A caused a significant reduction in shoot dry weight, protein and chlorophyll *a* and total contents. Notably, no significant difference was observed in the root growth, fresh dry weight and carbohydrate content by UV-R treatment. In addition, UV exposure resulted in a significant increase of flavonoid and anthocyanin contents.

**Keywords:** anthocyanin, flavonoid, chlorophyll, protein, carbohydrate.

## INTRODUCTION

radiation is the part of the non-ionizing region of the electromagnetic spectrum that comprises approximately 8%-9% of total solar radiation [6]. UV is traditionally divided into 3 wavelengths. UV-C (200-280 nm) is extremely harmful to living organisms, but not relevant under natural conditions of solar irradiation. UV-B (280-320 nm) is of particular interest because although this wavelength represents only approximately 1.5% of the total spectrum, it can have a variety of damaging effects in plants. UV-A (320-400 nm) represents approximately 6.3% of the incoming solar radiation and is the least hazardous part of UV radiation [8]. Projections indicate that solar UV-B radiation will reach peak levels on the surface of the earth within the next few years [13]. However, it is expected that UV-B radiation could fall to pre-ozone depletion levels by

2050 if the Montreal Protocol is fully implemented by all member countries [38]. UV radiation is readily absorbed by biomolecular such as amino acids, polypeptides and nucleic acids [8]. Enhanced UV radiation causes a reduction in plant growth and photosynthetic capacity [34] and pigment levels [29]. Increased UV exposure has been shown to alter the biotic relationships of higher plants, as demonstrated by the changes in plant disease susceptibility and the balance of competition between plant species. The influence of UV radiation on growth appears to be mediated by phytohormones, either via photodestruction or enzymatic reactions. Overall, the effects of UV radiation vary, both among species and among cultivars of a given species. Of those plants that have been tested, a large proportion exhibited reduced plant growth (plant height, dry weight, leaf area, etc.), photosynthetic activity, and flowering. Photosynthetic activity may be reduced by direct effects on the photosynthetic process or metabolic pathways, or indirectly through effects on photosynthetic pigments or stomatal function. Plants sensitive to UV may also respond by accumulating UV-absorbing compounds in their outer tissue layers, which presumably protect sensitive targets from UV damage. The key enzymes in biosynthetic pathways of these compounds have been shown to be specifically induced by UV irradiation via gene activation [35]. UV radiation above ambient levels may inhibit plant growth, development, reproduction, and photosynthesis [11, 29, 34]. However, plant sensitivity to UV radiation differs between species [32] and even varieties [1, 3, 28]. It is modified by the plant growth rate [5], developmental stage [33], growth form (herbs cf. trees), and functional type [7]. Additionally, air temperature [20], atmospheric carbon dioxide concentrations [30], and soil nitrogen [4, 10], phosphorus [23], and moisture [31] content may affect plant sensitivity to UV radiation [24]. The aim of the present study was to screen a variety of parameters considered to play an important role in plant protection against UV radiation to get an encompassing view of the way that important crop plants stand when exposed to enhanced levels of UV radiation.

## MATERIALS AND METHODS

### Plant growth and treatment

This study was conducted in biology department of Urmia university from 2010/04 to 2010/08. Savory, *Satureja hortensis L.*, is a member of the Lamiaceae family. Seeds of savory (obtained from natural resource of Urmia, Iran) were sterilized with 10% sodium hypochlorite for 10 min then soaked in distilled water. The percentage of germination was about 90%. The soil used in pots was obtained from a field and mixed with sand (1:5 v/v). The mixture was autoclaved at 121 °C for 4 h before use. The germinated seeds were grown in 45 pots measuring 20 cm in diameter in a greenhouse. After 50 days of growth under uniform conditions they were divided into 3 sets of 15 pots. One set served as the control, another set received UV-C(245nm) radiation for 8 days, which was produced by a UV-C germicidal lamp (TUV/G30T8, Philips, Holland) that provided an irradiation dose of approximately  $17.2 \text{ kJ m}^{-2} \text{ d}^{-1}$ , and the third set was exposed to UV-A(360nm) radiation for 15 days, which was produced by 2 insecticide UV-A lamps (F20T9/BL, Hitachi, Japan) that produced an irradiation dose of approximately  $18.9 \text{ kJ m}^{-2} \text{ d}^{-1}$ . Plants were grown at 25/20 °C (day/night) 16 h of light and 8 h of dark, and were alternately watered with half-strength Hoagland solution and distilled water. Plant height, root length and fresh weight were measured immediately after removing the plants from the experimental field. Dry weight was determined after drying at 80°C for 24h. Leaves of all plants from each treatment were counted and average number of leaves per plant was calculated.

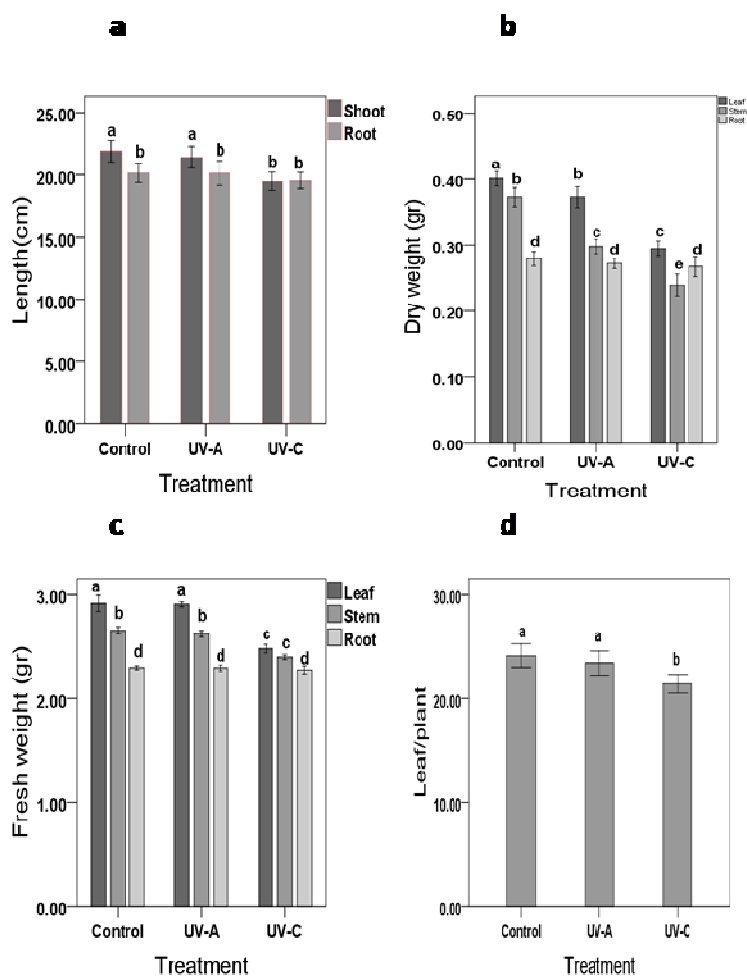


Fig., 1. Effect of UV-A and UV-C treatment on a) length of plants, b) dry weight, c) fresh weight, d) number of leaves, means followed by the same letters are not significantly difference at  $P < 0.05$  according to Tukey's test.

#### Flavonoid assay:

To determine the absorption by flavonoids, 0.1 gr of fresh leaf tissue were taken from the distal ends of an upper leaf and were extracted in 15 ml glass centrifuge tubes containing 10 ml ethyl alcohol: acetic acid (99:1 v:v). The samples were gently boiled for 10 min in a water bath at 80°C and brought up to volume absorbance was measured at three wavelengths: 270, 300, 330 nm with UV-vis spectrophotometer (model: LKB-Biochrom) [15].

#### Anthocyanin assay:

To determine the concentration of anthocyanin, 0.1 gr fresh leaves were taken and were extracted in 15 ml glass centrifuge tubes containing 10 ml of acidified methanol (methanol: HCL 99:1 v:v) and kept overnight in the dark. The samples were brought up to volume, and the absorbance at 550 nm determined. Anthocyanin concentration was calculated using an extinction coefficient of 33000  $\text{mol}^{-1}\text{cm}^{-1}$  [39].

**Protein assay:**

Protein concentration was evaluated by the method of Lowry et al, using bovine albumin serum as a standard [18].

**Pigments assay:**

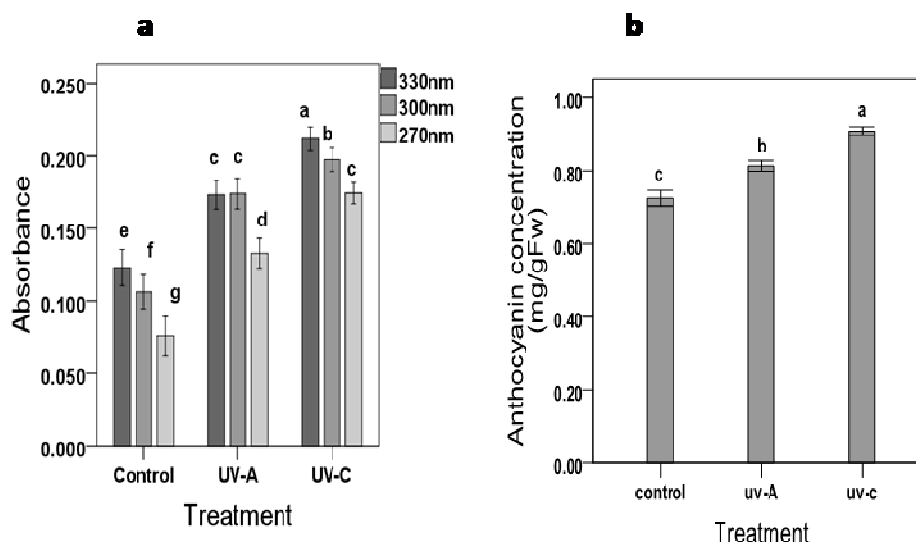
Chlorophylls and carotenoids were extracted from leaf discs with 80% acetone and determined according to Lichtenthaler (1987).

**Carbohydrate assay:**

Soluble and insoluble carbohydrates were determined by method of Fales (1951).

**Statistical analysis:**

Quantitative changes of different parameters were analysed through analysis of variance (Anova), with Tukey's multiple range test being used to determine significant differences among treatments.



**Fig., 2 Effect of UV-A and UV-C treatment on a) absorbance of the methanolic extract of leaves at 270, 300 and 330 nm, b) concentration of anthocyanin at 550 nm. Means followed by the same letters are not significantly different at  $P < 0.05$  according to Tukey's test.**

**RESULTS****Growth parameters**

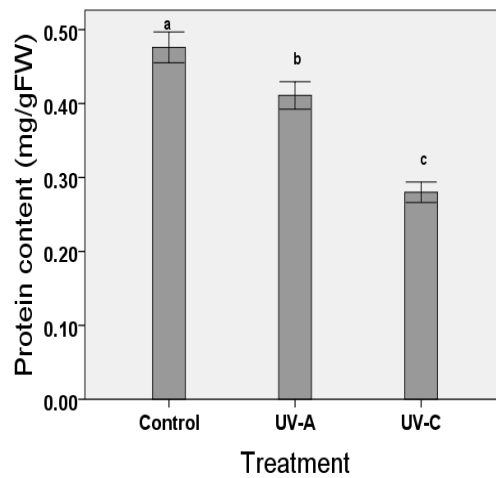
In comparison with the control plants, root length was not significantly changed by UV exposure, but shoot length decreased and this reduction was just significant in UV-C exposed plants (Fig., 1a). Shoot dry weight decreased in both UV-A and UV-C treated plants (Fig., 1b). Shoot fresh weight and total number of leaves decreased significantly in UV-C exposed plants (Fig., 1c-1d) but root fresh and dry weights did not show an appreciable change in UV-R exposed plants (Fig., 1b-1c).

**UV absorbing compounds**

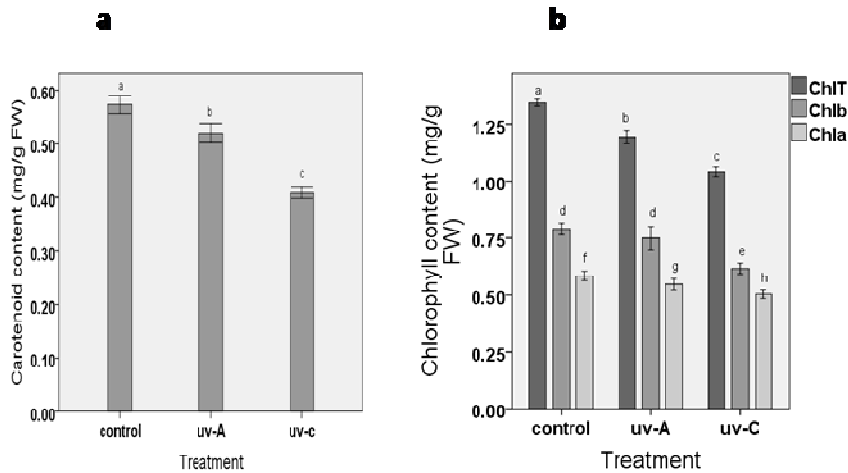
In this survey we found that exposure to UV radiation caused an increase in the UV absorbing compounds. Concentration of flavonoids has been significantly increased in both UV-A and UV-C treatments in comparison with control (Fig., 2a). Anthocyanin concentration has also been increased in UV-R exposed plants (Fig., 2b).

**Protein content:**

In this experiment we observed a significant decrease in leaf protein contents in both UV-A and UV-C treated plants (Fig.,3)



**Fig., 3 Effect of UV-A and UV-C treatment on Protein content, means followed by the same letters are not significantly difference at P< 0.05 according to Tukey’s test.**



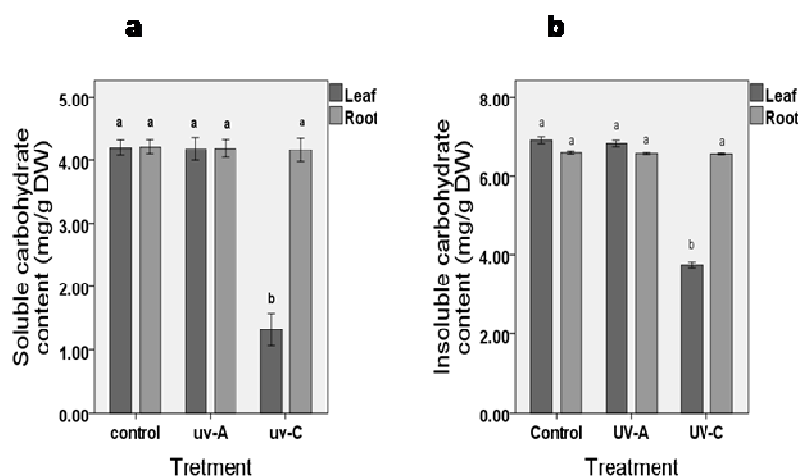
**Fig., 4 Effect of UV-A and UV-C treatment a)Carotenoid content, b)Chlorophyll content means followed by the same letters are not significantly difference at P< 0.05 according to Tukey’s test.**

**Pigment content:**

The content of chlorophyll-a, chlorophyll-b and chlorophyll-T (a + b) decreased in UV-R exposed. Only the reduction of chlorophyll-b in UV-A treated plants was not significant (Fig., 4b). Carotenoid concentration was also reduced in UV-R exposed plants and this reduction was significant (Fig., 4a).

**Carbohydrate content:**

The carbohydrate content was significantly decreased in shoot of UV-C treated plants but in UV-A treated plants there was no significant change in carbohydrate content. In the root of UV-R exposed plants no significant changes were observed in fresh weight (Fig., 5a-5b).



**Fig., 5 Effect of UV-A and UV-C treatment on a)soluble carbohydrate, b)Insoluble carbohydrate content, means followed by the same letters are not significantly difference at P < 0.05 according to Tukey's test.**

**DISCUSSION**

UV exposure decreased plant length and this decrease was significant in the UV-C-exposed plants. The growth of many species is reduced in response to UV treatment. Similar changes have been observed in *Capsicum longum* [9], *Fagopyrom tataricum* [40], *Pisum sativum* [26], strawberry [21] and sweet flag [16]. Since plant growth and development are closely related to the concentration of some endogenous plant growth regulators, such as IAA, therefore it is possible that the reduction in growth is a consequence of IAA reduction [36]. In this present study we found that UV-C decreased savory plants fresh weight. Similar changes have been observed in many species. In pea [12] and strawberry plants [21] also it has been shown that UV radiation exposure caused the reduction of biomass. Since photosynthesis is very important process in plant as it determines biomass increase [12] thus plants biomass reduction is related to inhibition of photosynthesis by UV radiation. UV-C radiation significantly reduced number of leaves per plant. Masih and kulkarnee (2010) have also reported reduction of leaf number under UV-B stress in strawberry plants [21]. In present research it is observed that savory plants responded to UV treatment by increasing their flavonoids and anthocyanin contents. Flavonoids found in a great variety of plant species, including *Capsicum annum* [19], *Arabidopsis thaliana* [17]. Flavonoids are induced and accumulated in plant tissues in response to UV irradiation. They can prevent UV-induced damage to plant tissues and plant with increased levels of flavonoids show

decreased sensitivity to damage by UV irradiation [14]. It is reported that anthocyanins and flavonoids protect leaf cells from photo oxidative damage from excess light and UV radiation [2]. In the present study savory leaves responded to the UV treatment by decreasing their protein content. Inactivation of proteins and enzymes can be caused directly by UV photolysis of aromatic amino acids or disulfide groups if affected residues are included in the active site [8]. It was reported that UV radiation increased protein content in *Brassica napus* plants [25]. This increment may be related to defense proteins and enzymes which are probably synthesis during stress [22]. The experimental results showed that UV irradiance caused the reduction of the contents of chlorophyll and carotenoid of savory leaves. Similar changes have been observed in *Capsicum annuum* [19]. Pigments of the photosynthetic apparatus can be destroyed by UV radiations, with concomitant loss of photo synthetic capacity. The chloroplast was the first organelle to show injury response when irradiated with UV-B radiation. The reduction in carotenoid content may result either from inhibition of synthesis or from breakdown of the pigments. Since the carotenoids are involved in the light harvesting and protection of chlorophyll from photo oxidative destruction, any reduction in carotenoid could have serious consequences of chlorophyll pigments [27]. In this present work, UV-C reduced content of shoot carbohydrates. In most species, sucrose is the principal form of carbohydrate translocated throughout the plant by the phloem. Starch is an insoluble carbohydrate reserve that is present in almost all plants. Both starch and sucrose are synthesized from the triose phosphate that is generated by the calvin cycle. The main CO<sub>2</sub>-fixing enzyme in C<sub>3</sub> plants is Rubisco, and comprises it about 50% of the total soluble plant protein. Rubisco activity declines with UV radiation [8]. Therefore, we concluded that UV-C-induced reduction of carbohydrate content could be due to inactivation of the Rubisco.

### CONCLUSION

This study shows that savory plants are sensitive to UV-R and indicate the sensitivity of these plants to UV-C was more than UV-A radiation.

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