

Impact of seed priming with different UV rays on morphological and physio-biochemical attributes of pea (*Pisum sativum* L.)

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Abstract

In order to investigate the effect of priming and UV stress on pea (*Pisum sativum* L. cv. Dorian), a pot experiment was conducted as factorial based on completely randomized design with 22 treatments (11 type of seed priming and 2 UV stress conditions) and four replications at Razi University during 2014. The results showed that the impact of seed pre-treatment was significant on all traits studied. Also, effect of UV stress was significant on relative water content (RWC), maximum quantum yield of PSII, total chlorophyll (Chl total) content and hydrogen peroxide concentration (H₂O₂). Interaction between seed priming and UV stress was also significant ($p \leq 0.01$) for RWC, Chl total content and H₂O₂ concentration. Generally, the results indicated that UV stress has harmful effect on the pea plants. On the other hand, hydro-priming (HP) had a better effect on the morphological characteristics (stem length and fresh weight) and RWC, especially, under non-UV stress condition. But, HP for 12 h + UV-AB for 2 h and also HP for 11 h + UV-AB for 3 h showed the lowest Chl total content, maximum quantum yield of PSII, stem length and fresh weight of plant and also had the highest concentration of H₂O₂. Therefore, these two pre-treatments have a negative impact on the pea plant and their use is not recommended for the pre-treatment of seeds in pea.

Keywords: Maximum quantum yield of PSII; Pea (*Pisum sativum* L.); Relative water content; Seed pre-treatment; Ultraviolet rays

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Abbreviations: Reactive oxygen species (ROS), Fresh weight of plant (FWP), Stem length (SL), Relative water content (RWC), Fresh weight (FW), Dry weight (DW), Turgid weight (TW), Total chlorophyll (Chl total), Chlorophyll (Chl), Hydrogen peroxide (H₂O₂), Malondialdehyde (MDA), Duncan's multiple range test (DMRT), Ultraviolet radiation (UV), Photosystem II (PSII), Photosynthetically active photon flux density (PPFD).

Introduction

Crop plants in natural conditions are routinely affected by several stresses acting simultaneously and/or in sequence. One of these types of tensions is ultraviolet radiation (UV). Although UV absorbs a small amount of sunlight (about 8-9%) but is divided into three bands. UV-A radiation (320-400 nm), UV-B radiation (280-320 nm) and UV-C radiation (200-280 nm) have long been recognized as being potentially damaging to living organisms (Kumar and Pandey 2017). The production of reactive oxygen species (ROS) under UV-stress are due to the metabolic

disturbance (Jenkins 2009; Hideg *et al.* 2013). The resistance of genotypes to UV-B irradiation depends on the activation of protective mechanisms, such as UV-B filters, quenchers of ROS, antioxidant enzymes and some metabolites of the Asada-Haliwell and xanthophyll cycles (Asada 1999; Bjorn *et al.* 2002; Caldwell *et al.* 2003; Lidon and Ramalho 2011; Lidon *et al.* 2012). Kargar Khorrami *et al.* (2013) reported that UV radiation, especially UV-B and UV-C, significantly decreased leaf surface, root and stem length, fresh and dry weight, photosynthetic pigments, protein and carbohydrate content in okra (*Hibiscus esculentus* L.). Also, Pourakbar and Abedzadeh (2014) showed that UV-B and UV-C rays reduced root and shoot weight of lemon balm (*Melissa officinalis*).

Methods for improving the performance and increasing the resistance of plants to environmental stresses, should be easy to operate and environmentally friendly. One of these is the use of seed priming techniques, such as HP and physical seed priming (e.g. ultraviolet radiation) (Yousefi and Fallah 2014; Dadrasi and Aboutalebian 2015; Rasaei *et al.* 2017). In experiments conducted on seeds of different legumes, including chickpea (*Cicer arietinum* L.), mung bean (*Vigna unguiculata* L.) and lentil (*Lens culinaris* Medik), the speed and percentage of germination and resistance to environmental stresses increased in the primed seeds (Kaur *et al.* 2006; Posmyk and Janas 2007; Ghassemi-Golezani *et al.* 2008). On the other hand, Khakpur *et al.* (2011) reported that treatment of seeds with ultraviolet radiation, especially UV-B, did not increase the rate of germination, root and plant

growth of flax varieties (*Linum usitatissimum*). Furthermore, Rasaei *et al.* (2017) observed that pre-treatment of seeds with water (HP) and HP + UV-A radiation had a positive effect on the growth and physiological characteristics of pea, but HP + UV-AB radiation was not effective on this plant.

In general, there is little information about the effect of priming by different bands of ultraviolet radiation. Therefore, the aim of this research was to determine the effect of seed priming (by water and different bands of ultraviolet radiation) and/or the effect of UV radiation alone on dry matter accumulation as well as physiological characteristics such as relative water content (RWC), maximum quantum yield of PSII and total chlorophyll (Chl total) content of leaves in pea (*Pisum sativum* L.).

Materials and Methods

Plant materials and treatments

In order to investigate the impacts of seed pre-treatment and UV stress on morphological and physio-biochemical characteristics of pea (*P. sativum* L. cv. Dorian), a pot experiment was conducted as factorial based on completely randomized design with 22 treatments (eleven types of seed priming and two conditions of UV stress) and four replications at Razi University, Kermanshah, Iran during 2014. The first factor consisted of 11 levels of seed priming as (1) control (non-seed priming), (2) HP for 14 h, (3) HP for 13 h and UV-A for 1 h, (4) HP for 12 h and UV-A for 2 h, (5) HP for 11 h and UV-A for 3 h, (6) HP for 13 h and UV-AB for 1 h, (7) HP for 12 h and UV-AB for 2 h, (8) HP for 11 h and

UV-AB for 3 h, (9) HP for 13 h and UV-C for 1 h, (10) HP for 12 h and UV-C for 2 h and (11) HP for 11 h and UV-C for 3 h. Seeds of the Dorian variety were obtained from Isfahan Agricultural and Natural Resources Research Center. The details of the method of pre-treatment are shown in Table 1. After drying the treated seeds at room temperature within 48 h, they were planted in pots. The pots having a diameter of 20 cm and height of 30 cm, filled with the mixture of perlite and coco peat with 2:1 ratio. Three seeds were sown in each pot with equal distances. The environmental condition of the hydroponic system was set to 14 hours of light and 10 hours of darkness, ambient temperature of 25/22 °C

(day/night), relative humidity of 55-65% and photosynthetically active photon flux density (PPFD) or brightness intensity of 180 $\mu\text{mol m}^{-2} \text{s}^{-1}$. The plants were fed twice a week with liquid fertilizer (Fosamco brand). For this purpose, 2 mL of the fertilizer was dissolved in 1 L of water and consumed. The second factor was two UV stress conditions including (i) no UV stress and (ii) UV radiation stress. UV stress was induced in an irradiation chamber with two narrow-band fluorescent lamps made in Germany (LT 18W/009 UV) at 21 days after sowing. After 35 days of culture, the physiological and morphological attributes were measured.

Table 1. Details of the priming treatments evaluated in this study and their code.

No.	Priming treatments	Code
1	Control (or non-priming)	C
2	Hydro-priming 14 h	HP
3	Hydro-priming 13 h + UV-A 1 h	HP + UV-A ₁
4	Hydro-priming 12 h + UV-A 2 h	HP + UV-A ₂
5	Hydro-priming 11 h + UV-A 3 h	HP + UV-A ₃
6	Hydro-priming 13 h + UV-AB 1 h	HP + UV-AB ₁
7	Hydro-priming 12 h + UV-AB 2 h	HP + UV-AB ₂
8	Hydro-priming 11 h + UV-AB 3 h	HP + UV-AB ₃
9	Hydro-priming 13 h + UV-C 1 h	HP + UV-C ₁
10	Hydro-priming 12 h + UV-C 2 h	HP + UV-C ₂
11	Hydro-priming 11 h + UV-C 3 h	HP + UV-C ₃

Relative water content

Leaf RWC was determined according to Bandurska (2000) for each UV stress treatments. RWC content was measured in the morning from 8:00 am to 10:00 am. For this purpose, leaf samples (500 mg) were soaked in 50 mL distilled water for 22 h at 4°C in the dusk and then their

turgid weight was recorded. After this time, they were oven-dried at 70 °C for 34 h and their dry weights was measured. RWC was computed as follows:

$$\text{RWC (\%)} = [(\text{FW} - \text{DW}) / (\text{TW} - \text{DW})] \times 100,$$
where FW, DW and TW are fresh weight, dry weight and turgid weight, respectively.

Maximum quantum efficiency of PSII

Maximum quantum efficiency of PSII was recorded on the two youngest leaves of each plant by using the time-resolving portable fluorimeter (PEA, Hansatech Instrument, Kings Lynn, UK). Leaf clips were placed on the leaves 20 min prior to the measurement to provide dark adaptation. After that, samples were illuminated with continuous red light (the peak wavelength of 650 nm, spectral line half-width of 22 nm). The light was provided by an array of three light-emitting diodes. The light pulse intensity was $3500 \mu\text{mol m}^{-2} \text{s}^{-1}$ and the duration of the light pulse was 2 s. Measurements were performed on the middle part of a leaf blade, away from the main leaf vein. Maximum quantum yield of PSII shows the maximal quantum yield of photochemistry in the dark-adapted state (Strasser *et al.* 1999). Maximum quantum yield of PSII was calculated according to the following equation:

$$\text{Maximum quantum yield of PSII} = (F_m - F_0) / F_m$$

where, F_m represent maximal fluorescence yield of dark-adapted sample with all PSII centers closed, and F_0 represent minimal fluorescence yield of dark-adapted sample with all PSII centers open.

Chlorophyll content

The fully expanded flag leaves on the stated dates were homogenized in ice cold 80% acetone (1.5 mL for a 250 mg sample) and extracted for 24 hours. Samples were centrifuged at $6000 \times g$ for 15 min at 4°C , then, the supernatants were collected. The pigment composition was measured using a double-beam spectrophotometer according to the method described by Lichtenthaler and Wellburn

(1983). This method involves measurement of the light absorbed in the leaf extract at 645 and 663 nm using an Elisa spectrophotometer (BioTek, PowerWave, USA).

$$\text{Chl a (mg/g FW)} = [(12.7 \times A_{663}) - (2.6 \times A_{645})] \times \text{mL acetone mg}^{-1}$$

$$\text{Chl b (mg/g FW)} = [(22.9 \times A_{645}) - (4.68 \times A_{663})] \times \text{mL acetone mg}^{-1}$$

$$\text{Chl total (mg/g FW)} = \text{Chl a} + \text{Chl b}$$

Hydrogen peroxide (H₂O₂) concentration

In order to determine the of H₂O₂ concentration, 0.3 g fresh mass of fully developed leaves were homogenized in a mortar at 4°C with 3 mL of 0.1% trichloroacetic acid and centrifuged for 20 min at $15000 \times g$. The reaction mixture contained 500 μL of the supernatant, 500 μL phosphate buffer with the pH of 7.4 and after adding 1 mL of 1 M KI, samples were incubated in the dark for 60 min and absorption was measured at $\lambda = 390 \text{ nm}$. The H₂O₂ content was calculated using a standard curve in the range of 1-100 nM/mL H₂O₂ (Jessup *et al.* 1994).

Stem length and plant fresh weight

Plants were harvested after 35 days of sowing, and their stem length and fresh weight were measured.

Statistical analysis

Before analysis of variance, all data sets were checked for normality of distribution and equality of variances. Analysis of variance was done using the SAS 8.0 statistical package. Means were compared by Duncan's multiple range test (Duncan 1955) at $p \leq 0.05$.

Results

The results showed that the effect of seed priming was significant on fresh weight of plant, stem length, RWC, maximum quantum yield of PSII, Chl total and H₂O₂ (Table 2). Also, the effect of

UV stress was significant on RWC, maximum quantum yield of PSII, Chl total and H₂O₂ (Table 2). Furthermore, significant interaction between seed priming and UV stress for RWC, Chl total and H₂O₂ concentration was observed (Table 2).

Table 2. Analysis of variance of the effect of seed priming and UV stress on fresh weight of plant, stem length, relative water content, maximum quantum yield of PSII, total chlorophyll and hydrogen peroxide concentration in pea (*Pisum sativum*).

Sources of variation	df	Mean squares					
		Fresh weight of plant	Stem length	Relative water content	Maximum quantum yield of PSII	Total chlorophyll	Hydrogen peroxide
Priming	10	57.8 **	215 **	0.544 **	0.007 **	0.545 **	32.1 **
UV	1	0.395 ns	3.36 ns	1751 **	0.034 **	20.5 **	1386 **
Priming × UV	10	0.067 ns	1.71 ns	0.356 **	0.003 ns	0.286 **	4.52 **
Error	66	0.344	2.03	0.110	0.002	0.051	0.089
CV (%)	-	6.69	4.17	0.35	5.94	7.69	1.29

ns, * and **, not significant and significant at the 5% and 1% levels of probability, respectively.

Morphological characteristics

Fresh weight of plant increased significantly as compared to the control, following of HP treatment. But, this trait decreased significantly following HP + UV-AB₂ and HP + UV-AB₃ treatments as compared to the control (Table 3). Similar trend was observed for stem length. HP treatment showed the highest and HP + UV-AB₃ showed the lowest stem length (40.2 cm and 23.2 cm, respectively) (Table 3). It seems that increasing the duration of UV radiation for seed pre-treatment has adverse effect on the measured morphological traits.

Physiological characteristics

The trend of the RWC in leaves of pea is shown in Table 4. RWC decreased (9.15%) in the plants

grown under UV stress as compared to the control condition (Table 4). Highest amount of RWC was observed in HP and HP + UV-C₂ treatments under non-UV stress condition (97.6%). However, the lowest amount of RWC was obtained in HP + UV-A₂ and HP + UV-AB₁ treatments under UV stress condition (87.9 and 87.8%, respectively) (Table 4). The highest reduction in RWC after exposure to UV stress was seen in HP + UV-AB₁ and HP + UV-A₃ (9.9 and 9.6%) treatments and the lowest reduction was seen in HP + UV-A₁ (8.3%) (Table 4).

HP + UV-C₁ treated plants had the highest and control (non-priming), HP, HP + UV-A₁, HP + UV-AB₂ and HP + UV-AB₃ treatments had the lowest maximum quantum yield of PSII, respectively (Table 3). Also, UV irradiation stress caused a significant decrease in maximum

quantum yield of PSII as compared to the control condition (no UV stress) (Table 5).

The results showed that the UV radiation stress causes the decline in photosynthetic

pigments of pea leaves on the average of priming treatments (Table 4). Higher amounts of Chl total was obtained for control (non-priming), HP, HP + UV-A₁, HP + UV-A₂, HP + UV-A₃, HP + UV-

Table 3. Influence of seed priming on fresh weight of plant, stem length and maximum quantum yield of PSII of pea (*Pisum sativum*).

Priming treatments	Fresh weight of plant (g)	Stem length (cm)	Maximum quantum yield of PSII
C	6.6 e	31.5 d	0.69 c
HP	11.6 a	40.2 a	0.70 c
HP + UV-A ₁	11.0 b	38.5 b	0.70 c
HP + UV-A ₂	11.1 ab	38.8 ab	0.76 ab
HP + UV-A ₃	11.5 ab	37.6 b	0.72 bc
HP + UV-AB ₁	8.4 d	32.7 d	0.76 ab
HP + UV-AB ₂	5.4 f	27.9 e	0.68 c
HP + UV-AB ₃	3.5 g	23.2 f	0.68 c
HP + UV-C ₁	10.3 c	37.8 b	0.77 a
HP + UV-C ₂	8.4 d	34.3 c	0.72 bc
HP + UV-C ₃	8.6 d	34.1 c	0.72 bc

Control or non-priming (C), hydro-priming for 14 h (HP), hydro-priming for 13 h and UV-A for 1 h (HP + UV-A₁), hydro-priming for 12 h and UV-A for 2 h (HP + UV-A₂), hydro-priming for 11 h and UV-A for 3 h (HP + UV-A₃), hydro-priming for 13 h and UV-AB for 1 h (HP + UV-AB₁), hydro-priming for 12 h and UV-AB for 2 h (HP + UV-AB₂), hydro-priming for 11 h and UV-AB for 3 h (HP + UV-AB₃), hydro-priming for 13 h and UV-C for 1 h (HP + UV-C₁), hydro-priming for 12 h and UV-C for 2 h (HP + UV-C₂), hydro-priming for 11 h and UV-C for 3 h (HP + UV-C₃).

Means in the same column followed by the same letter are not significantly different at $P \leq 0.05$ according to Duncan's multiple range test.

AB₁, HP + UV-C₁, HP + UV-C₂ and HP + UV-C₃ treatments under non-UV stress condition (Table 4). On the other hand, lower amounts of Chl total (1.80 and 1.90 mg/g FW) were recorded in HP + UV-AB₂ and HP + UV-AB₃ treatments, respectively under UV stress (Table 4). Furthermore, highest and lowest reduction in Chl total after exposure to UV stress was seen in HP + UV-AB₂ (46.7%) and HP + UV-A₃ (3.4%) treatments, respectively (Table 4). The concentration of H₂O₂ increased significantly (41.1%) in the leaves of pea plants grown at UV stress as compared to non-UV stress condition (Table 4). Higher amounts of H₂O₂ concentration were observed for HP + UV-AB₂ and HP + UV-

AB₃ treatments under UV stress condition (28.9 and 29.3 μ M/g FW, respectively). However, the lowest amount of H₂O₂ with 16.4 μ M/g FW belonged to HP + UV-C₁ treatment under non-UV stress (Table 4). Also, highest and lowest increase in the concentration of H₂O₂ after exposure to UV stress was seen in HP + UV-AB₁ (49.7%) and HP + UV-AB₂ (22.5%) treatments, respectively (Table 4).

Discussion

In our study, with applying UV stress, the amount of fresh weight of plant and stem length did not change significantly in the pea plants (Table 3). The results obtained in this study are inconsistent

with the results of Liu *et al.* (2013). They reported that UV stress reduced plant height, dry weight of individual stem, yield per plant, pod number per plant and seed number per pod (Liu *et al.* 2013). Also, UV radiation has been found to suppress plant growth and decrease germination in Kentucky bluegrass (Ervin *et al.* 2004).

Under UV stress in the laboratory, RWC and Chl total in pea plant decreased, but concentration of H₂O₂ increased as compared to non-UV condition (Table 4). In concordance with the results of this study, the reduction of RWC by UV stress has been reported by other researchers in wheat (He *et al.* 2011) and barley seedlings (Bandurska *et al.* 2012). Furthermore, many studies have reported the adverse effects of UV radiation on plant growth and physiological characteristics such growth retardation (Heijde and Ulm 2012; Bandurska *et al.* 2013), cell membrane degradation, destruction of chloroplasts and reduction of chlorophyll content (Lidon and Ramalho 2011; Lidon *et al.* 2012), reduced photosynthetic activity (Lidon and Ramalho 2011), and subsequent decrease in dry matter production in plants (Hideg *et al.* 2013). Hajihosseini *et al.* (2016) by studying two pumpkin genotypes under UV radiation indicated that root and shoot length, fresh and dry weight of root and shoot, leaf area, number of leaves per plant, leaf relative water content, content of chlorophyll a, b and carotenoids, decreased by the UV radiation stress as compared to the non-UV control. The causes of reduced chlorophyll content against UV rays include: (i) the destruction of pigment precursors, (ii) the inhibition of chlorophyll synthesis (Rasaei *et al.*

2017) and (iii) increased levels of ethylene (Zhang and Kirkham 1996; Severo *et al.* 2015).

UV irradiation stress caused a significant decrease in maximum quantum yield of PSII as compared to the control condition (no ultraviolet tension) (Table 5). The results obtained in this study were in agreement with those of Lidon and Ramalho (2011) in rice. Our results showed an oxidative stress due to UV radiation. The concentration of H₂O₂ increased in the leaves of pea plants grown under UV stress (Table 4). Generally, an unfavorable environment (including UV radiation) causes oxidative stress in plants which provokes formation of ROS (Gill and Tuteja 2010). Usually ROS lead to increased concentrations of MDA and H₂O₂ (Katerova *et al.* 2012) and proline content (Kapchina-Toteva *et al.* 2004), which are sensitive to stress. Pourakbar and Abedzadeh (2014) also reported an increase in the amount of H₂O₂ and MDA by UV rays (especially UV-B and UV-C) and stated that this result was due to the decrease in the activity of catalase enzyme.

Seed priming plays an important regulatory role in plants' growth and development. The present study showed that the seed priming by water and UV-A, UV-AB and UV-C lights affected the fresh weight of plant and stem length (Table 3) as well as physiological and biochemical characteristics (Table 4). As was indicated above, fresh weight of plant and stem length increased significantly following of HP and HP + UV-A pre-treatments, as compared to the control treatment. But, these traits decreased significantly following the pre-treatment of HP + UV-AB₂ and HP + UV-AB₃ treatments (Table 3).

Table 4. Influence of seed priming and UV stress on relative water content, total chlorophyll content and hydrogen peroxide concentration of pea (*Pisum sativum*).

Priming treatment	UV condition			Percentage change of each seed pre-treatment after UV stress (%)
	Non-stress	UV stress	Mean	
Relative water content (%)				
C	97.3 ^{ab}	88.1 ^{fgh}	92.7 ^{cde}	-9.5
HP	97.6 ^a	89.0 ^{cd}	93.3 ^a	-8.8
HP + UV-A ₁	97.2 ^{ab}	89.1 ^c	93.1 ^{ab}	-8.3
HP + UV-A ₂	96.9 ^b	87.9 ^h	92.4 ^e	-9.3
HP + UV-A ₃	97.3 ^{ab}	88.0 ^{gh}	92.6 ^{de}	-9.6
HP + UV-AB ₁	97.4 ^{ab}	87.8 ^h	92.6 ^{de}	-9.9
HP + UV-AB ₂	97.3 ^{ab}	88.5 ^{defg}	92.9 ^{abcd}	-9.0
HP + UV-AB ₃	97.3 ^{ab}	88.3 ^{efgh}	92.8 ^{bcde}	-9.2
HP + UV-C ₁	97.3 ^{ab}	88.6 ^{cdef}	92.9 ^{abcd}	-8.9
HP + UV-C ₂	97.6 ^a	88.5 ^{defg}	93.1 ^{ab}	-9.3
HP + UV-C ₃	97.3 ^{ab}	88.8 ^{cde}	93.0 ^{abc}	-8.7
Mean	97.3 ^a	88.4 ^b		
Total chlorophyll (mg/g FW)				
C	3.48 ^a	2.35 ^d	2.91 ^c	-32.5
HP	3.50 ^a	2.48 ^d	2.99 ^{bc}	-29.1
HP + UV-A ₁	3.48 ^a	2.50 ^d	2.99 ^{bc}	-28.2
HP + UV-A ₂	3.43 ^a	2.38 ^d	2.90 ^c	-30.6
HP + UV-A ₃	3.50 ^a	3.38 ^{ab}	3.44 ^a	-3.4
HP + UV-AB ₁	3.43 ^a	2.95 ^c	3.19 ^b	-14.0
HP + UV-AB ₂	3.38 ^{ab}	1.80 ^e	2.59 ^d	-46.7
HP + UV-AB ₃	3.05 ^{bc}	1.90 ^e	2.48 ^d	-37.7
HP + UV-C ₁	3.50 ^a	2.55 ^d	3.03 ^{bc}	-27.1
HP + UV-C ₂	3.48 ^a	2.48 ^d	2.98 ^{bc}	-28.7
HP + UV-C ₃	3.68 ^a	2.50 ^d	3.09 ^{bc}	-32.1
Mean	3.44 ^a	2.48 ^b		
Hydrogen peroxide (uM/g FW)				
C	18.9 ^h	27.4 ^c	23.1 ^c	45.0
HP	19.0 ^h	28.1 ^b	23.5 ^b	47.9
HP + UV-A ₁	19.0 ^h	28.1 ^b	23.6 ^b	47.9
HP + UV-A ₂	19.0 ^h	28.1 ^b	23.5 ^b	47.9
HP + UV-A ₃	18.6 ^{hi}	27.3 ^c	23.0 ^c	46.8
HP + UV-AB ₁	18.3 ^{ij}	27.4 ^c	22.8 ^c	49.7
HP + UV-AB ₂	23.6 ^f	28.9 ^a	26.2 ^a	22.5
HP + UV-AB ₃	23.4 ^f	29.3 ^a	26.3 ^a	25.2
HP + UV-C ₁	16.4 ^l	22.1 ^g	19.2 ^f	34.8
HP + UV-C ₂	17.1 ^k	25.3 ^e	21.2 ^e	48.0
HP + UV-C ₃	17.9 ^j	26.5 ^d	22.2 ^d	48.0
Mean	19.2 ^b	27.1 ^a		

Control or non-priming (C), hydro-priming for 14 h (HP), hydro-priming for 13 h and UV-A for 1 h (HP + UV-A₁), hydro-priming for 12 h and UV-A for 2 h (HP + UV-A₂), hydro-priming for 11 h and UV-A for 3 h (HP + UV-A₃), hydro-priming for 13 h and UV-AB for 1 h (HP + UV-AB₁), hydro-priming for 12 h and UV-AB for 2 h (HP + UV-AB₂), hydro-priming for 11 h and UV-AB for 3 h (HP + UV-AB₃), hydro-priming for 13 h and UV-C for 1 h (HP + UV-C₁), hydro-priming for 12 h and UV-C for 2 h (HP + UV-C₂), hydro-priming for 11 h and UV-C for 3 h (HP + UV-C₃). Means followed by different letters in the same column or row and also in the two columns consisting of the combination of priming treatments and UV conditions, are significantly different at $p \leq 0.05$ according to Duncan's multiple range test.

Table 5. Influence of UV stress on maximum quantum yield of PSII of pea (*Pisum sativum*).

UV condition	Maximum quantum yield of PSII
Non-stress	0.73 a
UV stress	0.70 b

Means in the same column followed by different letter are significantly different at $p \leq 0.05$ according to Duncan's multiple range test.

The results of this study indicate that low doses of UV radiation (e.g. UV-A), probably, act as a germination stimulant and improve the growth process. However, high energy UV rays (especially UV-B and UV-C) have inhibitory effects on germination and plant growth. It can be argued that UV ray (especially UV-AB) damages cell membranes, which results in an increase in the concentration of H_2O_2 (Table 4) and ultimately the reduction of biomass (Table 3). Mahdavian *et al.* (2006) in pepper (*Capsicum annuum* L.) and Pourakbar and Abedzadeh (2014) in lemon balm (*Melissa officinalis* L.) reported that UV-B and UV-C rays reduced root and shoot weight. Also, Kargar Khorrami *et al.* (2013) showed that UV-B and UV-C had serious effects on okra plant, but UV-A was not harmful to this species.

Conclusion

In conclusion, the results of this study showed that UV stress negatively affected physiological and morphological attributes of pea plant at vegetative stage. Pre-treatment of seeds with HP had a positive effect on pea. However, the combination of HP with UV radiation was less efficient than

HP alone. Especially, treatments HP + UV-AB₂ and HP + UV-AB₃ had the lowest Chl total content, maximum quantum yield of PSII, stem length and fresh weight and also highest concentration of hydrogen peroxide. Therefore, these pre-treatments (HP + UV-AB₂ and HP + UV-AB₃) have a negative impact on the pea plant and their use is not recommended for the pre-treatment of pea seeds.

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اثر پرایمینگ بذر با اشعه‌های مختلف فرابنفش بر پارامترهای مورفولوژیکی و فیزیولوژیکی-بیوشیمیایی نخود (*Pisum sativum* L.) فرنگی

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چکیده

به منظور ارزیابی تاثیر پرایمینگ و تنش فرابنفش بر رقم دورین نخود فرنگی (*Pisum sativum* L.)، یک آزمایش گلدانی به صورت فاکتوریل بر پایه طرح کاملا تصادفی با ۲۲ تیمار (ترکیب ۱۱ نوع پرایمینگ بذر و ۲ شرایط تنش فرابنفش) در چهار تکرار در دانشگاه رازی طی سال ۱۳۹۴ به اجرا درآمد. نتایج نشان داد که تاثیر پیش تیمارهای بذر بر تمام صفات مورد مطالعه معنی‌دار است. همچنین اثر تنش فرابنفش بر محتوای آب نسبی، حداکثر عملکرد کوانتومی فتوسیستم II، کلروفیل کل و غلظت پراکسید هیدروژن معنی‌دار بود. اثر متقابل پرایمینگ بذر با تنش فرابنفش بر محتوای آب نسبی، کلروفیل کل و غلظت پراکسید هیدروژن در سطح احتمال یک درصد معنی‌دار بود. به طور کلی نتایج نشان داد که تنش فرابنفش اثر مخربی بر روی گیاه نخود فرنگی می‌گذارد. از سوی دیگر، هیدروپرایمینگ تاثیر بهتری بر صفات مورفولوژیکی (ارتفاع ساقه، وزن تر بوته) و فیزیولوژیکی (محتوای آب نسبی برگ) این گیاه، به ویژه در شرایط بدون تنش فرابنفش، داشت. اما ترکیب‌های تیماری هیدروپرایمینگ به مدت ۱۲ ساعت + UV-AB به مدت ۲ ساعت و نیز هیدروپرایمینگ به مدت ۱۱ ساعت + UV-AB به مدت ۳ ساعت از کمترین میزان کلروفیل کل، حداکثر عملکرد کوانتومی فتوسیستم II، ارتفاع ساقه و وزن تر بوته و بیشترین غلظت پراکسید هیدروژن برخوردار شدند. در نتیجه، این پیش تیمارها تاثیر منفی بر گیاه نخود فرنگی دارند و استفاده از آن‌ها برای پیش تیمار بذر نخود فرنگی توصیه نمی‌شود.

واژه‌های کلیدی: اشعه‌های فرابنفش؛ پیش تیمار بذر؛ حداکثر عملکرد کوانتومی فتوسیستم II؛ نخود فرنگی (*Pisum sativum* L.)؛ محتوای آب نسبی