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Study of phytohormones effects on UV-B stress seeds of thyme species

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ABSTRACT

Background & Aim: *Thymus vulgaris* L. and *Thymus daenensis* Celak (Thyme), members of the family Lamiaceae, are widely used in Iranian folk medicine. The aim of this research was to study how salicylic acid (SA), gibberellin (GA), and indole acetic acid (IAA)-seed priming affect UV-B radiation in seeds of Thyme (*T. vulgaris*, *T. daenensis* Celak) under both laboratory and outdoor conditions.

Experimental: The effects of phytohormones (200, 400, 600 ppm) and irradiation performed in a 60 Co Gamma cell 220 source at a dose of 3 kGy (10, 20 and 30 min) on seeds of Thyme species were studied. Seeds were tested under *in vitro* and outdoor conditions in a complete randomized factorial layout with 4 and 3 replications, respectively. The characters measured under *in vitro* condition were seed germination percentage, mean time of germination, root and stem length. On the field, the characters evaluated were number of leaf, length of plant, root and shoot dry matter and essential oil.

Results: SA treatment was better under *in vitro* conditions. No significant effects were obtained from GA and IAA. The most destructive effects and the best beneficial phytohormones were UV 30 min and SA (200 and 400 ppm), respectively. The effects of foliar application of hormones were determined on growth and essential oil production in medicinal plants in two consecutive years. Shoot dry matter increased in both years with SA applications but IAA and GA had no beneficial effects on germination characters after UV radiation.

Recommended applications/industries: SA-priming of seeds protects thyme seedlings against UV-B radiation. The results of this study also showed that elevation of free SA levels in plants, either by exogenous feeding or genetically may enhance their tolerance to abiotic stress.

1. Introduction

Thymus species are commonly used as flavoring agents and medicinal plants. Seeds of this plant do not exhibit dormancy and the best time for harvesting this plant is at the beginning of its flowering period (Yadegari, 2015).

During the past decades, the thinning of the stratospheric ozone has led to enhanced ultraviolet-B (UV-B) radiation on the earthsurface. UV-B radiation (280-320 nm) represents only 0.3% of the radiation

reaching the Earth, but it is significant since it is absorbed by important macromolecules, including proteins and nucleic acids (Caldwell *et al.*, 2007). Increases in solar UV-B have raised concerns about the damaging impact of UV-B radiation on crop plants (Taipina *et al.*, 2011). Numerous studies have shown that enhanced UV-B radiation can affect the physiological and biochemical processes of many plant species, including altered plant photosynthesis (Sunita and Guruprasad, 2012), changes in carbon partitioning from growth pools to secondary metabolic pathways (Liu *et al.*, 2013), and thus changes in crop morphology, crop reproductive organ abortion and yield reduction (Choudhary and Agrawal, 2014), plant growth and development, photosynthesis and biomass production (Lavola *et al.*, 2013; Nawkar *et al.*, 2013). UV stress might influence the ratio of seed materials such as total anthocyanin (Eguchi and Sato, 2009), phenolic compounds (Kondo and Kawashima, 2000), as well as malondialdehyde, superoxide dismutase, catalase and peroxidase (Chen, 2008). Enhanced UV decreased the leaf area per unit plant biomass but increased biomass productivity (Weih *et al.*, 1998), had no significant effects on height growth, shoot and root biomass of birch seedlings (Kostina *et al.*, 2001), increased superoxide dismutase (SOD) activity (Li *et al.*, 2014), number of seeds per fruit as well as mean individual seed mass but the germination rates of produced seeds were unaffected (Stephanou and Manetas, 1998). In wheat, the combination of drought and UV-B caused more severe damage to wheat seedlings than stress factors applied separately (Tian and Lei, 2007). UV induces fluorescence of cotton foreign matter (Mustafic *et al.*, 2014). Responses to high energy UV-B cause noticeably different effects in different varieties of plants. If plant experience higher energy UV in the early stages of growth, the effects can cause heritable changes, which will in turn affect yield and impact the viability of the next generation of seed (Brown and Jenkins, 2008). Application of UV-B light can be used as a strategy to improve antioxidant phenolic compounds of plants (Lee *et al.*, 2013).

Plant hormones play important roles in plant growth and yield formation (Xu and Li, 2006). Salicylic acid (SA) is considered to be an important signaling molecule, which plays an important role in regulating a number of physiological processes and plant resistances to stresses (Saruhan *et al.*, 2012). Many reports have illustrated that SA can ameliorate the injurious effects of UV on plants (Moussaand El-Gamal, 2010; Liu *et al.*, 2012). SA seed priming reduced UV-B radiation on growth (Singh *et al.*, 2015), concentrations of several polyphenols and produced more total phenolic, flavonoids, and antioxidants (Lee *et al.*, 2013). Plants accumulate large amounts of SA when exposed to UV radiation (Bandurska and Cieślak, 2013). Furthermore, SA stimulates the photosynthetic machinery by increasing the content and accumulation of chlorophyll in UV-stressed plants (Mahdavian *et al.*, 2008) in response to oxidative stress. Although UV induced oxidative stress and increased lipid peroxidation that was significantly decreased by SA pre-treatment (Saruhan *et al.*, 2012). The application of SA to plants improved shoot growth and photosynthesis (Fattahi *et al.*, 2011). Depending on the plant species, the application of Gibberellin (GA), a phytohormone, can improve plant growth, ion uptake and transport, nutrient utilization and is responsible for seed germination, stem elongation, leaf expansion and flowering, and prevents chlorophyll breakdown (Rashad and Hussien, 2014).

Auxin is another phytohormone and was first isolated and characterized for plant growth. Indole-3-acetic acid (IAA) is a type of auxin (Nakamura *et al.*, 2006; Bilkay *et al.*, 2010). The application of Indole Acetic Acid (IAA) resulted in more starch accumulation and is thus an essential compound required for the growth and development of roots and shoots (Yang *et al.*, 2014).

The aim of this study was to investigate the effect of phytohormones and irradiation on measured characters such as speed of germination, mean time of germination, root and stem length under *in vitro* and outdoor conditions. Number of leaf, length of plant, root and shoot dry matter and essential oil were measured.

2. Materials and Methods

2.1. Plant material and treatments

The seeds of *T. vulgaris* and *T. daenensis* Celak were collected from the fields. The treatments were salicylic acid (SA), Gibberellin (GA), Indole Acetic Acid (IAA) and regulation of UV-B toxicity in seeds under both laboratory and outdoor conditions at the Islamic Azad University, Shahrekord Branch. In this research, seeds of thyme were studied to determine the effects of phytohormones (200, 400, 600 ppm) and irradiation was performed in a 60 Co Gamma cell 220 source at a dose of 3 kGy (10, 20 and 30 min). Under *in vitro* and outdoor conditions a complete randomized block design in factorial layout with 4 and 3 replications, respectively was used. Before medicinal plants culture, each petri was washed with alcohol, disinfected by fire and placed on a stain paper. After disinfection, seeds were treated with 2000 ppm Carboxin thiram at 25°C temperature, under light. When water in the petri dish was relatively evaporated, irrigation was conducted in the field. The number of seedling produced by each plant was counted every day. Measured characters under *in vitro* condition were seed germination percentage, mean time of germination, root and stem length. Number of leaf, length of plant, root and shoot dry matter and essential oil were measured. The germination ratio was measured with the following formula (Fattahi *et al*., 2011):

Speed of Germination (SP) = X1 /Y1 + (X2-X1)/Y2 + …. + (Xn-Xn-1)/Yn

Mean Time of Germination (MTG) = N1T1+N2T2+N3T3+ … (NnTn)/ N

Where N1, N2 are the number of seeds germinated on the first and second days, respectively; T1=N1, T2=N1+N2, N is the total seed germinated, Y_n number of days to final germination period.

Three replicates of the control and treated plants (three plants of each) were harvested and the shoot of each plant was collected separately for estimation of shoot fresh weight and height which were expressed as g/plant and cm/shoot, respectively. Irradiation was performed in a ^{60}Co Gamma cell 220 (AECL) source with a dose rate of about 3.5 kGy/h at doses of 3 kGy having a dose uniformity factor of 1.13. Dosimetry mapping was previously performed by Fricke dosimetry.

2.2. Essential oil extraction

Fresh aerial parts of *T. vulgaris* L. and *T. daenensis* Celak were dried at room temperature (25±3°C). Plant material was powdered (100 g) and subjected to hydro–distillation with 1000 ml distilled water for 3 h using a Clevenger-type apparatus.

2.3. Statistical analysis

All data were subjected to ANOVA using the statistical computer package SAS ver. 8. When the treatment effects were significant ($p<0.05$) in relation to the control, treatment means were separated using Least Significant Difference test at the significance level ($P<0.05$).

3. Results and discussion

IAA and GA hormones had no significant effects on seeds treated with UV. Results from the field showed that there was a significant difference between concentrations of SA. Although in some single treatments, there were no significant differences, but in combined treatments, there were differences in most characters as the SA400×UV10 treatment was the best (Tables 1 to 4). It seems that by increasing UV radiation, most of the measured characters were diminished. The application of SA treatment significantly improved growth characters including stem and root length, number of lateral stem/ leaf, fresh weight of root and shoot, dry weight of root and shoot and essential oil percentage in two species of thyme. The destructive effects of UV on seeds under laboratory conditions of the two species were in appearance; only SA could repair morphological measured characters.

Table 1. L.S.D comparisons of measured characters of *Thymus vulgaris* L. that affected by SA and UV in first year under field condition.

Treat	SL[†] (cm)	RL (cm)	MTG	SP (%)	FWR (g/plant)	FWS (g/plant)	DWR (g/plant)	DWS (g/plant)	EO (%)	NLS	NL
M1[¥]	6.7±0.1^{a}	5.9±0.1^{a}	8.1±1.1^{a}	0.18±0.01^{a}	95.4±5.1^{a}	111.1±8.1^{a}	35.2±3.1^{a}	41.1±5.3^{a}	0.8±0.01^{a}	11.1±1.1^{a}	99.2±3.3^{a}
M2	7.2±0.1^{a}	6.1±0.06^{a}	8.2±0.9^{a}	0.19±0.02^{a}	96.3±4.2^{a}	117.5±7.1^{a}	36.3±4.2^{a}	45.2±4.3^{a}	0.8±0.01^{a}	11.4±2.2^{a}	99.4±4.2^{a}
M3	4.3±0.09^{b}	4.4±0.1^{b}	7.4±0.8^{a}	0.17±0.01^{a}	92.1±3.1^{a}	98.9±6.7^{ab}	33.1±3.2^{a}	39.4±2.1^{a}	0.8±0.01^{a}	10.7±1.3^{a}	90.4±6.1^{a}
M4	5.5±0.08^{ab}	5.5±0.05^{ab}	6.6±0.7^{b}	0.15±0.01^{b}	85.4±5.2^{a}	94.2±4.1^{b}	27.2±4.2^{ab}	34.3±3.2^{ab}	0.6±0.01^{b}	9.5±1.4^{a}	73.2±5.2^{b}
M5	5.3±0.08^{ab}	5.2±0.1^{ab}	6.7±0.6^{b}	0.16±0.01^{b}	86.1±3.3^{a}	97.1±5.7^{ab}	28.2±3.2^{ab}	35.7±2.7^{ab}	0.6±0.02^{b}	9.6±2.2^{a}	74.2±6.3^{b}
M6	3.3±0.09^{c}	3.2±0.05^{b}	5.9±1.1^{b}	0.12±0.01^{b}	72.1±4.1^{b}	88.1±4.1^{b}	25.6±2.2^{ab}	8.2±4.5^{b}	0.5±0.01^{b}	8.8±1.1^{a}	65.1±5.3^{b}
M7	1.3±0.1^{d}	1.2±0.04^{c}	4.8±1.1^{c}	0.09±0.01^{c}	61.1±5.5^{b}	72.2±3.3^{c}	22.3±2.1^{ab}	25.2±4.3^{b}	0.2±0.01^{c}	4.5±1.7^{b}	26.4±1.2^{c}
M8	1.4±0.09^{d}	1.4±0.1^{c}	4.9±0.5^{c}	0.1±0.06^{c}	62.2±5.5^{b}	75.2±4.2^{c}	23.3±3.1^{ab}	25.5±6.1^{b}	0.2±0.01^{c}	4.4±0.9^{b}	27.2±1.2^{c}
M9	1.1±0.08^{d}	1±0.07^{c}	3.9±0.8^{c}	0.07±0.05^{c}	48.2±3.1^{c}	61.1±5.1^{c}	18.2±1.1^{b}	19.1±3.3^{c}	0.2±0.01^{c}	3.1±0.9^{b}	19.1±1.1^{c}

Means in each column followed by the same letters are not significantly different (P<0.05).

[†]SL: Stem length, RL: Root length, NLS: Number of lateral stem, NL: Number of leaf, FWR: Fresh weight of root, FWS: Fresh weight of shoot, DWR: Dry weight of root, DWS: Dry weight of shoot, EO: Essential oil, MTG: Mean Time of Germination, SP: SP: Speed of Germination.

[¥]M1: UV10×SA200, M2: UV10×SA400, M3: UV10×SA600, M4: UV20×SA200, M5: UV20×SA400, M6: UV20×SA600, M7: UV30×SA200, M8: UV30×SA400, M9: UV30×SA600.

Table 2. L.S.D comparisons of measured characters of *Thymus vulgaris* L. that affected by SA and UV in second year under field condition.

Treat	SL[†] (cm)	RL (cm)	MTG	SP (%)	FWR (g/plant)	FWS (g/plant)	DWR (g/plant)	DWS (g/plant)	EO (%)	NLS	NL
M1[¥]	7.7±0.1^{a}	5.8±0.05^{a}	8.5±1.1^{a}	0.17±0.01^{a}	88.2±2.1^{a}	114.4±2.1^{a}	30.2±3.2^{a}	39.2±1.1^{a}	0.72±0.01^{a}	12.4 ±1.1^{a}	93.2 ±5.5^{a}
M2	7.4±0.2^{a}	6.2±0.04^{a}	8.5±1.2^{a}	0.18±0.01^{a}	89.3±3.2^{a}	121.5±3.4^{a}	32.1±2.1^{a}	41.1±2.3^{a}	0.81±0.01^{a}	14.2 ±1.8^{a}	96.7 ±3.1^{a}
M3	5.3±0.2^{b}	4.7±0.03^{b}	7.2±1.3^{a}	0.11±0.01^{b}	72.1±2.1^{a}	98.2±4.4^{ab}	28.2±2.2^{a}	35.3±1.1^{a} ^{b}	0.74±0.01^{a}	11.7 ±2.1^{a}	96.4 ±3.3^{a}
M4	5.7±0.3^{ab}	5.7±0.04^{a} ^{b}	6.4±0.5^{b}	0.1±0.01^{b}	82.2±3.2^{a}	92.2±3.5^{b}	29.1±1.1^{a}	34.3±2.4^{a} ^{b}	0.49±0.01^{b}	9.7± 1.1^{b}	75.4 ±2.1^{b}
M5	4.8±0.1^{ab}	5.5±0.1^{ab}	6.4±0.6^{b}	0.1±0.01^{b}	71.1±4.1^{a}	90.3±2.2^{b}	32.2±2.1^{a}	35.7±3.1^{a} ^{b}	0.51±0.01^{b}	9.4± 0.2^{b}	75.2 ±2.1^{b}
M6	3.7±0.2^{c}	3.1±0.02^{b}	5.8±0.7^{b}	0.09±0.01^{b}	72.1±5.5^{a}	70.2±3.3^{c}	28.6±1.2^{a}	27.1±2.6^{b}	0.47±0.01^{b}	8.6± 0.2^{b}	65.6 ±2.2^{b}
M7	0.99±0.3^{d}	1.3±0.01^{c}	4.9±0.1^{c}	0.09±0.01^{b}	59.1±3.1^{b}	67.7±2.4^{c}	15.4±2.1^{b}	24.2±2.1^{b}	0.26±0.02^{c}	4.3± 0.31^{c}	25.3 ±2.1^{c}
M8	0.95±0.2^{d}	1.4±0.01^{c}	4.9±0.8^{c}	0.08±0.01^{b}	54.2±2.2^{b}	55.2±3.2^{d}	15.5±2.2^{b}	25.2±1.2^{b}	0.25±0.02^{c}	4.9± 0.2^{c}	22.4 ±2.3^{c}
M9	0.87±0.3^{d}	0.99±0.0 1^{c}	3.6±0.1^{c}	0.05±0.01^{c}	44.9±2.4^{b}	45.1±2.3^{d}	11.1±2.3^{b}	18.2±1.1^{c}	0.19±0.02^{c}	3.7± 0.1^{c}	16.5 ±2.7^{c}

Means in each column followed by the same letters are not significantly different (P<0.05).

[†]SL: Stem length, RL: Root length, NLS: Number of lateral stem, NL: Number of leaf, FWR: Fresh weight of root, FWS: Fresh weight of shoot, DWR: Dry weight of root, DWS: Dry weight of shoot, EO: Essential oil, MTG: Mean Time of Germination, SP: SP: Speed of Germination.

[¥]M1: UV10×SA200, M2: UV10×SA400, M3: UV10×SA600, M4: UV20×SA200, M5: UV20×SA400, M6: UV20×SA600, M7: UV30×SA200, M8: UV30×SA400, M9: UV30×SA600.

Table 3. L.S.D comparisons of measured characters of *Thymus daenensis* L. that affected by SA and UV in first year under field condition.

Treat	SL[†] (cm)	RL (cm)	MTG	SP (%)	FWR (g/plant)	FWS (g/plant)	DWR (g/plant)	DWS (g/plant)	EO (%)	NLS	NL
M1	10.2±0.7^{a}	7.6±0.5^{a}	10.1±1.2^{a}	0.21±0.02^{a}	81.2±3.2^{a}	99.4±3.2^{a}	22.2±0.6^{a}	29.3±1.2^{a}	0.62±0.02^{a}	11.4±1.1^{a}	83.2±0.6^{a}
M2	10.7±0.8^{a}	8.7±0.2^{a}	10.5±0.9^{a}	0.22±0.01^{a}	82.3±2.4^{a}	111.1±4.2^{a}	21.1±0.6^{a}	31.1±1.4^{a}	0.7±0.03^{a}	12.2±1.3^{a}	86.7±0.4^{a}
M3	8.4±0.7^{b}	6.3±0.1^{b}	9.2±0.8^{a}	0.17±0.01^{b}	73.1±2.2^{a}	98.6±3.2^{a}	20.2±0.5^{a}	25.3±2.1^{a}	0.66±0.01^{a}	10.7±1.2^{a}	86.4±0.4^{a}
M4	6.8±0.5^{b}	5.4±0.1^{b}	7.4±0.7^{b}	0.15±0.01^{b}	62.2±2.2^{b}	82.2±2.1^{b}	19.1±0.5^{a}	24.3±1.1^{a}	0.44±0.01^{b}	8.7±0.9^{b}	75.4±0.1^{b}
M5	7.3±0.4^{b}	5.7±0.2^{b}	7.7±0.6^{b}	0.14±0.02^{b}	66.1±3.1^{b}	88.3±3.2^{b}	18.2±0.1^{a}	25.7±0.8^{a}	0.51±0.01^{b}	8.4±0.8^{b}	75.2±0.1^{b}
M6	5.4±0.3^{c}	5.1±0.2^{b}	5.8±0.7^{b}	0.11±0.01^{c}	58.1±1.1^{b}	81.2±1.1^{b}	16.6±0.1^{b}	22.1±0.9^{b}	0.43±0.02^{b}	7.6±0.6^{b}	55.6±0.1^{c}
M7	3.6±0.1^{c}	2.3±0.2^{c}	4.9±0.1^{c}	0.09±0.01^{c}	49.1±3.2^{c}	57.7±2.1^{c}	14.4±0.6^{b}	19.2±0.7^{b}	0.22±0.02^{c}	4.2±0.5^{c}	15.3±0.5^{d}
M8	3.9±0.1^{c}	3.4±0.1^{c}	4.9±0.1^{c}	0.07±0.01^{c}	44.2±2.2^{c}	54.2±2.2^{c}	14.5±0.5^{b}	18.2±0.4^{b}	0.21±0.01^{c}	4.5±0.4^{c}	12.4±0.8^{d}
M9	2.8±0.2^{c}	2.9±0.1^{c}	4.6±0.3^{c}	0.04±0.01^{d}	34.9±2.1^{c}	44.1±2.7^{c}	13.1±1.1^{c}	15.2±0.1^{b}	0.17±0.01^{c}	3.7±0.6^{c}	11.5±0.9^{d}

Means in each column followed by the same letters are not significantly different (P<0.05).

[†]SL: Stem length, RL: Root length, NLS: Number of lateral stem, NL: Number of leaf, FWR: Fresh weight of root, FWS: Fresh weight of shoot, DWR: Dry weight of root, DWS: Dry weight of shoot, EO: Essential oil, MTG: Mean Time of Germination, SP: SP: Speed of Germination.

[¥]M1: UV10×SA200, M2: UV10×SA400, M3: UV10×SA600, M4: UV20×SA200, M5: UV20×SA400, M6: UV20×SA600, M7: UV30×SA200, M8: UV30×SA400, M9: UV30×SA600.

Table 4. L.S.D comparisons of measured characters of *Thymus daenensis* L. that affected by SA and UV in second year under field condition.

Treat	SL[†] (cm)	RL (cm)	MTG	SP (%)	FWR (g/plant)	FWS (g/plant)	DWR (g/plant)	DWS (g/plant)	EO (%)	NLS	NL
M1[¥]	11.7±1.2^{a}	7.8±0.4^{a}	9.5±0.5^{a}	0.22±0.01^{a}	78.2±2.2^{a}	99.4±1.6^{a}	22.2±1.3^{a}	35.2±0.9^{a}	0.65 ±0.02^{a}	12.7 ±0.3^{a}	85.2 ±2.5^{a}
M2	12.4±1.1^{a}	7.2±0.5^{a}	9.9±0.6^{a}	0.23±0.01^{a}	79.3±3.2^{a}	111.5±1.1^{a}	23.1±1.1^{a}	40.1±0.8^{a}	0.66 ±0.01^{a}	14.6 ±0.2^{a}	86.7 ±3.6^{a}
M3	10.3±0.9^{a}	6.7±0.6^{a}	8.2±0.3^{a}	0.2±0.01^{a}	76.1±3.3^{a}	88.2±1.1^{ab}	21.2±1.2^{a}	32.3±0.7^{ab}	0.59 ±0.01^{a}	11.5 ±0.1^{a}	81.4 ±4.1^{a}
M4	7.7±0.8^{b}	5.2±0.4^{b}	6.4±0.2^{b}	0.17±0.02^{b}	62.2±1.1^{ab}	72.2±1.1^{b}	19.1±1.4^{ab}	29.3±0.8^{ab}	0.47 ±0.01^{b}	8.7± 0.1^{b}	65.4 ±2.1^{b}
M5	7.8±0.7^{b}	4.6±0.1^{b}	6.4±0.4^{b}	0.16±0.02^{b}	66.1±1.2^{ab}	75.3±1.1^{b}	18.2±1.1^{ab}	31.7±0.9^{ab}	0.48 ±0.01^{b}	8.8± 0.1^{b}	66.2 ±2.1^{b}
M6	6.7±0.1^{b}	4.3±0.1^{b}	5.8±0.4^{b}	0.15±0.03^{b}	59.1±2.6^{ab}	66.2±2.3^{c}	15.6±1.3^{ab}	28.1±0.9^{b}	0.39 ±0.01^{b}	7.8± 0.1^{b}	67.6 ±2.7^{b}
M7	3.9±0.6^{c}	4.9±0.3^{b}	5.9±0.1^{b}	0.14±0.01^{b}	44.1±2.9^{c}	52.7±2.5^{c}	11.4±1.6^{b}	19.2±0.9^{b}	0.17 ±0.03^{c}	4.7± 0.3^{c}	17.3 ±3.1^{c}
M8	3.6±0.5^{c}	4.4±0.3^{b}	5.9±0.1^{b}	0.12±0.01^{c}	412±1.1^{c}	55.2±2.1^{c}	11.5±1.1^{b}	19.2±1.2^{b}	0.18 ±0.04^{c}	4.9± 0.3^{c}	18.4 ±1.1^{c}
M9	2.8±0.1^{c}	3.1±0.4^{c}	3.6±0.5^{c}	0.11±0.01^{c}	34.9±1.5^{c}	44.1±2.7^{c}	9.1±1.2^{b}	17.2±1.3^{c}	0.15 ±0.05^{c}	3.5± 0.2^{c}	16.5 ±1.9^{c}

Means in each column followed by the same letters are not significantly different (P<0.05).

†SL: Stem length, RL: Root length, MTG: MTG: Mean Time of Germination, SP: SP: Speed of Germination, FWR: Fresh weight of root, FWS: Fresh weight of shoot, DWR: Dry weight of root, DWS: Dry weight of shoot, EO: Essential oil, NLS: Number of lateral stem, NL: Number of leaf.
¥M1: UV10×SA200, M2: UV10×SA400, M3: UV10×SA600, M4: UV20×SA200, M5: UV20×SA400, M6: UV20×SA600, M7: UV30×SA200, M8: UV30×SA400, M9: UV30×SA600.

Table 5. Results of correlation between characters in *Thymus vulgaris* L. that affected by SA and UV in first year under field condition.

Characters	SL† (1)	RL (2)	NLS (3)	NL (4)	FWR (5)	FWS (6)	DWR (7)	DWS (8)	EO (9)	MTG (10)	SP (11)
1	1										
2	0.49*	1									
3	0.68**	0.57**	1								
4	0.78**	0.48*	0.5**	1							
5	0.6**	0.78**	0.45*	0.5**	1						
6	0.65**	0.76**	0.65**	0.59*	0.6**	1					
7	0.79**	0.65**	0.65**	0.65**	0.6**	0.65**	1				
8	0.68**	0.74**	0.69**	0.69**	0.72**	0.69**	0.66**	1			
9	0.88**	0.78**	0.73**	0.66**	0.77**	0.73**	0.71**	0.73**	1		
10	0.82**	0.71**	0.78**	0.75**	0.72**	0.74**	0.67**	0.55**	0.73**	1	
11	0.81**	0.73**	0.66**	0.71**	0.73**	0.67**	0.66**	0.8**	0.57**	0.66**	1

ns,* and ** : Non significant, significant at the 5% and 1% levels of probability, respectively.
†SL: Stem length, RL: Root length, NLS: Number of lateral stem, NL: Number of leaf, FWR: Fresh weight of root, FWS: Fresh weight of shoot, DWR: Dry weight of root, DWS: Dry weight of shoot, EO: Essential oil, MTG: Mean Time of Germination, SP: SP: Speed of Germination.

Table 6. Results of correlation between characters in *Thymus vulgaris* L. that affected by SA and UV in second year under field condition

Characters	SL† (1)	RL (2)	NLS (3)	NL (4)	FWR (5)	FWS (6)	DWR (7)	DWS (8)	EO (9)	MTG (10)	SP (11)
1	1										
2	0.49*	1									
3	0.68**	0.37ns	1								
4	0.78**	0.2 ns	0.55**	1							
5	0.6**	0.68**	0.48*	0.3 ns	1						
6	0.65**	0.66**	0.61**	0.51*	0.55**	1					
7	0.79**	0.61**	0.65**	0.55**	0.34 ns	0.61**	1				
8	0.68**	0.74**	0.65**	0.62**	0.65**	0.62**	0.62**	1			
9	0.88**	0.75**	0.81**	0.77**	0.71**	0.76**	0.66**	0.55**	1		
10	0.77**	0.7**	0.71**	0.55**	0.66**	0.71**	0.6**	0.5**	0.8**	1	
11	0.61**	0.66**	0.55**	0.66**	0.7**	0.62**	0.55**	0.72**	0.5**	0.6**	1

ns,* and ** : Non significant, significant at the 5% and 1% levels of probability, respectively.
†SL: Stem length, RL: Root length, NLS: Number of lateral stem, NL: Number of leaf, FWR: Fresh weight of root, FWS: Fresh weight of shoot, DWR: Dry weight of root, DWS: Dry weight of shoot, EO: Essential oil, MTG: Mean Time of Germination, SP: SP: Speed of Germination.

Table 7. Results of correlation between characters in *Thymus daenensis* L. that affected by SA and UV in first year under field condition.

Characters	SL† (1)	RL (2)	NLS (3)	NL (4)	FWR (5)	FWS (6)	DWR (7)	DWS (8)	EO (9)	MTG (10)	SP (11)
1	1										
2	0.33 ^{ns}	1									
3	0.62**	0.33 ^{ns}	1								
4	0.71**	0.38 ^{ns}	0.35 ^{ns}	1							
5	0.62**	0.58**	0.33 ^{ns}	0.44 ^{ns}	1						
6	0.55**	0.56**	0.56**	0.59*	0.6**	1					
7	0.65**	0.35 ^{ns}	0.61**	0.65**	0.6**	0.65**	1				
8	0.6**	0.54**	0.6**	0.65**	0.7**	0.63**	0.61**	1			
9	0.66**	0.5**	0.7**	0.8**	0.66**	0.53**	0.55**	0.8**	1		
10	0.55**	0.66**	0.45**	0.58**	0.44*	0.35^{ns}	0.45**	0.48**	0.8**	1	
11	0.34^{ns}	0.65**	0.37^{ns}	0.68**	0.35^{ns}	0.55**	0.3^{ns}	0.4^{ns}	0.88**	0.8**	1

ns,* and ** : Non significant, significant at the 5% and 1% levels of probability, respectively.

†SL: Stem length, RL: Root length, NLS: Number of lateral stem, NL: Number of leaf, FWR: Fresh weight of root, FWS: Fresh weight of shoot, DWR: Dry weight of root, DWS: Dry weight of shoot, EO: Essential oil, MTG: Mean Time of Germination, SP: SP: Speed of Germination.

Table 8. Results of correlation between characters in *Thymus daenensis* L. that affected by SA and UV in second year under field condition.

Characters	SL† (1)	RL (2)	NLS (3)	NL (4)	FWR (5)	FWS (6)	DWR (7)	DWS (8)	EO (9)	MTG (10)	SP (11)
1	1										
2	0.49*	1									
3	0.68**	0.57**	1								
4	0.78**	0.48*	0.5**	1							
5	0.6**	0.78**	0.45*	0.3 ^{ns}	1						
6	0.65**	0.76**	0.65**	0.3 ^{ns}	0.56**	1					
7	0.79**	0.65**	0.65**	0.55**	0.62**	0.65**	1				
8	0.68**	0.74**	0.69**	0.61**	0.62**	0.59**	0.62**	1			
9	0.88**	0.78**	0.83**	0.72**	0.73**	0.55**	0.66**	0.71**	1		
10	0.81**	0.7**	0.73**	0.7**	0.6**	0.5**	0.6**	0.7**	0.57**	1	
11	0.82**	0.7**	0.7**	0.66**	0.61**	0.75**	0.76**	0.55**	0.59**	0.59**	1

ns,* and ** : Non significant, significant at the 5% and 1% levels of probability, respectively.

†SL: Stem length, RL: Root length, NLS: Number of lateral stem, NL: Number of leaf, FWR: Fresh weight of root, FWS: Fresh weight of shoot, DWR: Dry weight of root, DWS: Dry weight of shoot, EO: Essential oil, MTG: Mean Time of Germination, SP: SP: Speed of Germination.

Thus, the foliar application of SA in 400 ppm concentration resulted in an increase which when compared with other treatments.

SA was found to be the most effective resisting the severe UV effects on its seeds. In almost the same order, the destructive effects of UV on the seeds could be reduced. UV radiation may be controlled but to a limited extent by SA. MTG as well as the SP ratio of the seeds, significantly increased as affected by SA and decreased upon non-application of SA. Stem length, root length, number of lateral stem/ leaf, fresh weight of root and shoot, dry weight of root and shoot,

essential oil percentage were measured on the two species of thyme in control. Most of the measured characters decreased upon application of more than 10 minutes of UV. Control treatment in many places produced the same yield components with SA400×UV10 treatment. It was clear from the presented data that the higher levels of UV were more effective than the lower levels, and SA hormone was superior to other hormones. However, the highest essential oil percentage was found with the SA400×UV10 treatment. Essential oil percentage positively correlated with the weight of shoot/root dry and fresh matter in the two species (tables 5-8). The results of this study showed just only the beneficial effects of SA hormone application on thyme plants. Increasing UV can result in lesser yield of essential oil content from thyme. In the present study, the effects of hormones applications were determined on the growth and yield of medicinal plants in two consecutive years. Shoot dry matter increased in both years with SA applications but IAA and GA had no beneficial effects on seeds after UV radiation.

Enhanced UV radiation has been found to suppress plant growth, decrease germination and growth (Ervin *et al.*, 2004). The highest germination rate was observed with SA (Fattahi *et al.*, 2011). SA can ameliorate oxidative stressed plants and the application of 400 ppm SA was beneficial and after this dosage, the expression of characters decreased. Under UV conditions in the laboratory, essential oil, yield and phytochemical materials in plant decreased (Eguchi and Sato, 2009; Weih *et al.*, 1998; Kondo and Kawashima 2000; Chen, 2008), or increased total phenolic, flavonoids, antioxidants (Lee et al., 2013), number of seeds per fruit (Stephanou and Manetas, 1998). Enhanced UV-B reduced plant height, dry weight of individual stem, yield per plant, pod number per plant and seed number per pod (Liu *et al*., 2013), concentrations of several polyphenols and had positive impacts on germination. Increasing UV-B radiation may have had no significant effects on the height growth, shoot and root biomass of birch seedlings (Kostina *et al*., 2001) or might alter biosynthesis of SA as indicated by a decreased level of SA (Singh *et al.*, 2015; Tian and Lei, 2007; Mustafic *et al.*, 2014). Plant responses to high energy UV-B cause noticeably different effects in different varieties of plants (Brown and Jenkins, 2008). SA pre-treatment significantly decreased UV induced oxidative stress and also acts as a stabilizer of membrane integrity to improve plant resistance to UV (Li *et al.*, 2014; Taipina *et al*., 2011).

This is the first report in which the effects of phytohormones applications on certain physiological characteristics were determined for seeds of thyme plants and also their relationship with yield and essential oil content. It is obvious that weight and number of stem can be used as a tool for selecting new cultivars with high yield under UV-B radiation. It seems the weight of shoot dry matter was the best character correlated by most characters. From the present data, it is thus concluded that the application of SA is beneficial to thyme plants at concentrations of 400 ppm or lower, and can result in an increase in dry matter yield of up to 40%. There are still many unanswered questions about how SA acts in increasing yield and essential oil content. One possibility is that the foliar applied SA can affect dry matter accumulation and increase shoot/root dry matter; also, SA affected the absorption and transition of essential nutrients which changes metabolism, growth and development and thus increased upper phytochemicals. It seems that control plants (no foliar application) performed better than plants subjected to upper UV radiation. These results reflect the role of applying SA in improving the total essential oils in thyme plants. Plants treated with 400 ppm of SA resulted in more fresh and dry weight of shoots and roots and essential oil percentage. Many researchers have stated that SA is an important signaling molecule which plays an important role in regulating plant resistance to stresses (Saruhan *et al.*, 2012; Moussa and El-Gamal, 2010; Liu *et al.*, 2012; Bandurska and Cieślak, 2013; Mahdavian *et al.*, 2008). SA pre-treatment enhanced seed germination percentage, mean time of germination, root and stem length; and in outdoor conditions, number of leaf, length of plant, root and shoot dry matter and essential oil percentage in the two species of thyme plant (Fariduddin *et al.*, 2003; Li *et al.*, 2014).

4. Conclusion

In this research, the SA-priming of seeds protects thyme seedlings against UV-B radiation. The results of this study also point out that the elevation of free SA levels in plants, either by exogenous feeding or genetically may enhance their tolerance against abiotic stress. Furthermore, the findings are also significant as it has been shown that UV stress occurs in natural fields where one stressor may modify the effects of others and SA may play a positive role in regulating toxicity in plants. It could be concluded from the results that SA hormone had significant effects on measured characters as well as the chemical component of the essential oil of thyme plants. The most suitable amount of SA supply for the production of thyme plants to obtain the highest weight of dry/fresh root and shoot in Shahrekord, Iran is 400 ppm foliar application. The effects of the combined application of UV10 × SA400 is suggested in comparison with their individual use. This study provides some useful information about the efficacy of foliar application of phytohormones under *in vitro* and *in vivo* conditions. These methods are relatively new and need further improvement with regard to rates, timing, and technique.

5. References

Bandurska, H. and Cieślak, M. 2013. The interactive effect of water deficit and UV-B radiation on salicylic acid accumulation in barley roots and leaves. *Environmental of experimental and Botanical*, volume 94: 9-18.

Bilkay, I.S., Karakoç, S. and Aksöz, N. 2010. Indole-3-acetic acid and gibberellic acid production in *Aspergillus niger*. *Turkish Journal of Biology*, volume 34: 313-318.

Brown, B.A. and Jenkins, G.I. 2008. UV-B signaling pathways with different flounce-rate response profiles are distinguished in mature Arabidopsis leaf tissue by requirement for UVR8, HY5, and HYH. *Plant Physiology*, volume 146: 576- 588.

Caldwell, M.M., Bornman, J.F., Ballare, C.L. and Kulandaivelu, G. 2007. Terrestrial ecosystems, increased solar ultraviolet radiation and interactions with other climatic factors. *Photochemistry Photobiology Science*, volume 6: 252-266.

Chen, Y.P. 2008. *Isatis indigotica* seedlings derived from laser stimulated seeds showed improved resistance to elevated UV-B. *Plant Growth Regulators*, volume 55: 73–79.

Choudhary. K.K. and Agrawal, S.B. 2014. Ultraviolet-B induced changes in morphological, physiological and biochemical parameters of two cultivars of pea (*Pisum sativum* L.). *Ecotoxicology Environment Safety*, volume 100:178–187.

Eguchi, K. and Sato, T. 2009. Differences in the Ratios of Cyanidin-3-O-glucoside and Cyanidin-3-O-rutinocide to Total Anthocyanin under UV and Non-UV Conditions in Tartary Buckwheat (*Fagopyrum tataricum* Garten). *Plant Production Science*, volume 12 issue 2: 150―155.

Ervin, E.H., Zhang, X. andFike, J.H. 2004. Ultraviolet-B radiation damage on Kentucky bluegrass. II. Hormone supplement effects. *Horticultural Science*, volume 39: 1471-1474.

Fattahi, M., Nazeri, V., Sefidkon, F., Zamani, Z. and Palazon, J. 2011. The Effect of Pre-sowing Treatments and Light on Seed Germination of *Dracocephalum kotschyi* Boiss: An Endangered Medicinal Plant in Iran. *Horticultural Environmental Biotechnology*, volume 52 issue 6: 559-566.

Kostina, E., Wulff, A. andTiitto, R.J. 2001. Growth, structure, stomatal responses and secondary metabolites of birch seedlings (*Betula pendula*) under elevated UV-B radiation in the field. *Trees*, volume 15: 483–491.

Lavola, A., Nybakken, L., Rousi, M., Pusenius, J. and Petrelius, M. 2013. Combination treatment of elevated UVB radiation, CO2 and temperature has little effect on silver birch (*Betula pendula*) growth and phytochemistry. *Physiology Plant*, volume 149: 499–514.

Liu, B., Liu, X.B., Li, Y.S. and Herbert, S.J. 2013. Effects of enhanced UV-B radiation on seed growth characteristics and yield components in soybean. *Field Crop Research*, volume 154: 158-163.

Lee, M.J., Son, J.E. and Oh, M.M. 2013. Growth and phenolic content of Sowthistle grown in a closed-type plant production system with a UV-A or UV-B

Lamp. *Horticultural EnvironmentalBiotechnology*, volume 54 issue 6: 492-500.

Li, X.M., Ma, L.J., Bu, N., Li, Y.Y. and Zhang, L.H.2014. Effects of salicylic acid pre-treatment on cadmium and/or UV-B stress in soybean seedlings. *Biologia Plantarum*, volume 58 issue 1: 195-199.

Mahdavian, K., Kalantari, K.M., Ghorbanli, M. and Torkzade, M. 2008. The effects of salicylic acid on pigment contents in ultraviolet radiation stressed pepper plants. *Biology of Plant*, volume 52:170-172.

Moussa, H.R. and El-Gamal, S.M. 2010. Effect of salicylic acid pretreatment on cadmium toxicity in wheat. *Biology of Plant*, volume 54: 315-320.

Mustafic, A., Li, Ch. and Haidekker, M. 2014. Blue and UV LED-induced fluorescence in cotton foreign matter. *Journal of Biology Engineering*, volume 8 issue 29: 2-11.

Rashad, R.T. andHussien, R.A. 2014. A comparison study on the effect of some growth regulators on the nutrients content of maize plant under salinity conditions. *Annals of Agricultural Science*, volume 59 issue 1: 89-94.

Saruhan, N., Saglam, A. and Kadioglu, A. 2012. Salicylic acid pretreatment induces drought tolerance and delays leaf rolling by inducing antioxidant systems in maize genotypes. *Acta Physiology Planetarium*, volume 34: 97-106.

Singh, V.P., Kumar, J., Singh, M., Singh, S., Prasad, S.M. and Dwivedi, R. 2015. Role of salicylic acid-seed priming in the regulation of chromium (VI) and UV-B toxicity in maize seedlings. *Plant Growth Regulators*, DOI 10.1007/s10725-015-0076-4.

Sunita, K., and Guruprasad, K.N. 2012. Solar UV-B and UV-A/B exclusion effects onintra specific variations in crop growth and yield of wheat varieties. *Field Crop Research*, volume 125: 8–13.

Taipina, M.S., Garbelottib, M.L., Leda, C.A. and Rodas, A.B. 2011. The effect of gamma irradiation on the nutrional properties of sunflower whole grain cookies. *Procedia Food Science*, volume 1:1992-1996.

Tian, X.R. and Lei, Y.B. 2007. Physiological Responses of Wheat Seedlings to Drought and UV-B Radiation. Effect of Exogenous Sodium Nitroprusside Application. *Russian Journal of Plant Physiology*, volume 54 issue 5: 676–682.

Xu, Z.H. and Li, J.Y. 2006. Plant hormones research in China: past, present and future. *Chaining Bulletin of Botany*, volume 23: 433–442.

Yadegari, M. 2015. Foliar application of micronutrients on essential oils of borago, thyme and marigold. *Journal of Soil Science and Plant Nutrition*, volume 15 issue 4: 949-964.

Yang, D., Luo, Y., Ni, Y., Yin, Y., Yang, W., Peng, D., Cui, Z. and Wang, Z. 2014. Effects of exogenous ABA application on post-anthesis dry matter redistribution and grain starch accumulation of winter wheat with different stay green characteristics. *Crop Journal*, volume 2: 144-153.