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[Research]

Effect of water-deficit stress on secondary metabolites of *Melissa officinalis* L.: role of exogenous salicylic acid

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ABSTRACT

Melissa officinalis is a perennial herbaceous plant from Lamiaceae family, widely used in food and pharmaceutical industries to add aroma. Drought stress in plants may happen due to the increase in water loss, insufficient water absorption or both. Salicylic acid (SA) is an important secondary metabolite in plants with hormone-like action in some biochemical pathways. Adding it during water-deficit stress, may alter a number of physiological processes, increasing the resistance of plant against possible damage by water loss. The aim of this study was to examine the effect of drought stress (DRU) on secondary metabolites of *Melissa officinalis* and also the efficacy of salicylic acid treatment to minimize the damages. According to the results, treatment of plant with 0.7 mM salicylic acid had considerable alteration in the quantity of important secondary metabolites. Protein concentration increased in all stages; Chlorophyll a reached to the highest amount; the amount of the MDA increased at the first and second stages in 1/3 fc drought treatment as well as at the third stage in 2/3 fc. Besides, total phenol content increased at stage I in 1/3 fc drought and the flavonoid content increased in 1/3 fc drought at all stages. Finally, content of anthocyanin was increased at first and third stages 1/3 fc drought, as well as at second stage in 1/3 fc drought. In conclusion, application of salicylic acid (0.7 mM) was useful for improving quality of lemon balm subjected to stress due to water deficit.

Key words: Drought stress, salicylic acid, *Melissa officinalis* L., Photosynthetic pigments, antioxidant capacity.

INTRODUCTION

Lemon balm, a member of Lamiaceae family, is well known for its medicinal properties (Zargari 1990). It is also used traditionally in many countries, including Iran, to add an aromatic flavor to herbal preparations. In common with many other plants, lemon balm undergoes physiological alterations during a period of drought (Sharafzadeh *et al*. 2012).

A number of physiological aspects of the plant are altered during water stress. The adverse effect is especially important on photosynthetic capacity and hormonal action. In long term, the growth and productivity of the plant could significantly decrease. However, many plants have specific adaption mechanism to resist many types of environmental stresses. Low water stress causes a significant increase in the concentration of solutes, which promotes the osmotic flow of water out of the plant cells (Douglas 1993). Treatment of the plant during various growth stages with some useful exogenous phytochemicals is a scientific way to adapt various stresses. Salicylic acid (SA) is a secondary metabolite in many plants with a phenolic group in its chemical structure. The compound acts as a phytohormone regulating some physiological processes. It has been reported that binding of SA can alter the activity of some plant proteins (Klessig 2017). It is worth indicating that SA and its derivatives have also multiple protein targets in animals.

Some of these proteins, similar to their plant counterparts, are associated with pathological processes. Salicylic acid (SA) alters a number of metabolic pathways including synthesis, oxidation and some biological activities including respiration, photosynthesis and absorption (Borsani *et al.* 2001). Its use during a low water stress could modify many biochemical pathways leading to improved plant resistance to drought stress (Yazdanpanah *et al*. 2011). It has been explained that SA is involved in activation of the stress - induced antioxidant system if plants are exposed to stress (Huang *et al.* 2008). The present study was designed to investigate the effect of salicylic acid, as a plant hormone, to reduce the impact of drought stress on some important characteristics of *Melissa officinalis* L.

MATERIALS AND METHODS

Materials

The study was conducted in University of Guilan during 2016. A completely randomized program with 3 replications was designed to investigate the effect of salicylic acid on a number of physiological processes of lemon balm (*Melissa officinalis* L.) under conditions of drought stress. The drought levels of (0, 1/3 field capacity, 2/3 fc) were combined with salicylic acid (0, 0.7 and 1.5 mM) treatments throughout the study period. The media was composed of sand, clay and manure fertilizer with 3:1:1 ratio (Table 1).

Lemon balm seeds were obtained from Iranian Institute of Agricultural Plants, Rasht, Iran. All chemicals and solvents were of analytical grade and purchased from Sigma Chemical Company.

Table 1. Selected physical and chemical characteristics of the soil.

Soil texture	Ec (ds m^{-1})	pH	P (ppm)	K (ppm)	N (%)	Organic C (%)
Silty clay loam	0.298	7.52	85	460	0.14	1.01

Abbreviations: P, phosphorous, K, potassium, N, nitrogen and C, carbon.

Methods

Plant growth and harvesting

Seeds were firstly disinfected with formaldehyde (3%, v/v) for 4 minutes followed by washing with distilled water at least 5 times to prevent fungal infection. They were then planted in pots containing 3:1:1 of sand, clay and manure fertilizer, respectively. Harvesting of lemon balm was carried out in three stages, i.e. before (stage I), during (stage II) and after flowering (stage III).

Preparation of samples for scanning electron microscopy (SEM)

The plant material used for histochemical investigation of the length of trichomes, was taken from the fresh samples, as well as the samples preserved in Formalin-aceto-alcohol fixative (FAA) The fixed leaf fragments were dehydrated in increasing alcohol concentration, i.e. 15, 30, 50, 70, 90 and 99.5 %. The plant samples were kept for 15 minutes in each concentration, followed by freeze drying overnight. In the next step, the specimens were placed on the base of an electron microscope (Stub), were gold plated with a Spotter Coated (POLARON SC 7620) and finally, observed using an electron microscope (LEO 1430 VP).

Determination of pigments content

Total chlorophyll concentration is a unifying parameter to determine the effect of any specific interventions. In this investigation, the concentrations of total chlorophyll (Chl a, Chl b) and carotenoids were measured by spectrophotometric method (Lichtenthaler & Wellburn 1983). The chlorophylls a and b and carotenoids were extracted from 0.2 g of leaves using 80% (v/v) aqueous acetone and the mixture was centrifuged. Absorbance of Chl a, Chl b and carotenoids extracts were obtained at 663, 645 and 470 nm, respectively. The concentrations of pigments were calculated as (mg g^{-1} fresh weight) from the following relationships:

Chlorophyll a (µg ml^{-1}) = 12.21 (A663) - 2.81 (A646)

Chlorophyll b (µg ml^{-1}) = 20.13 (A646) - 5.03 (A663)

Total Chlorophyll = Chlorophyll a + Chlorophyll b
Carotenoids = [(1000 × OD 470 nm – 1.8 × Chlorophyll a – 85.02 × Chlorophyll b)/198]

Determination of protein content by Bradford method

100 mg of Coomassie Brilliant Blue G-250 dissolved in 50 ml of 95% ethanol and 100 ml of 85% (w/v) phosphoric acid, was added to the mixture. The solution was diluted to 1 liter until the dye was completely dissolved. The resulting mixture was filtered through Whatman#1 paper just before use. A standard curve was obtained by plotting absorbance versus micrograms of protein and used to calculate the concentration of protein considering the dilution factor (Bradford 1976).

Determination of malondialdehyde (MDA) content

Malondialdehyde (MDA) content was measured by the well-known thiobarbituric acid method with a slight modification (Heath & Packer 1968). In practice, 0.5 g of tissue was homogenized in 5 ml of 5% (w/v) trichloroacetic acid and the homogenate was centrifuged at 12000g for 15 minutes at room temperature. An equal volume of thiobarbituric acid (0.5% in 20% trichloroacetic acid w/v) was added to the supernatant and mixed well. The resulting mixture was boiled for 25 minutes at 100° C and centrifuged at 7500g for 5 min. Absorbance of the supernatant was then measured at 532 nm and corrected for non-specific turbidity by subtracting the absorbance at 600 nm. The content of MDA was calculated using an extinction coefficient of 155 $M^{-1}cm^{-1}$.

Determination of total phenol

The concentration of phenolics in plant extracts was determined using a spectrophotometric method (McDonald *et al.* 2001*)*. Practically, a methanolic solution of the extract (concentration 1 mg ml^{-1}) was used in the analysis procedure. The reaction mixture was prepared by mixing 100 µl of the extract solution, 2 ml of 2% of $NaHCO_3$, 100 µl of 10% Folin-Ciocalteu's reagent dissolved in water and, finally, 2.8 ml of distilled water. The samples were incubated in a thermostated bath for 45 min at 45 °C and the absorbance of solution was obtained at 720 nm. The samples were prepared in triplicate for each analysis and the mean value of absorbance was obtained.

A similar procedure was repeated for the standard solution of gallic acid and the calibration line was prepared. Based on the measured absorbance, the concentration of phenolic compounds was read (mg ml^{-1}) from the calibration line. This content in extracts was expressed in terms of gallic acid equivalent (mg of GA g^{-1} of extract).

Determination of flavonoid content

The flavonoids present in the plant extracts were determined using a known spectrophotometric method (Singleton *et al.* 1999). The examined sample was composed of 1.5 ml of the extract solution with a concentration of 50 µl, and 100 µl in 10 % $AlCl_3$, 100 µl potassium acetate and 2.8 ml distilled water in 80% methanol. The samples were initially incubated at room temperature for at least one hour and their 415 nm absorbances were then measured. All samples were prepared in triplicate for each analysis and the mean value of absorbance was calculated. The same procedure was repeated for the standard solution of rutin and the calibration line was obtained.

Using this calibration line, the concentration of flavonoids was calculated (mg ml^{-1}). The content of flavonoids in extracts was, therefore, expressed in terms of rutin equivalent (mg of RU g^{-1} of extract).

Determination of anthocyanins

To determine the concentration of anthocyanins, 200 mg of leaf was extracted in 3 ml acidified methanol (methanol: HCl, 99:1, v:v) and stored for overnight in a dark place. The absorbance of the resultant solution was determined at 550 nm and anthocyanin concentration was calculated considering the extinction coefficient of 330 $cm^{-1}mol^{-1}$ (Wagner & Wüthrich 1979).

Variance analysis

The experiment was arranged as a factorial in a completely randomized block design with three replications. Duncan test was employed for grouping of treatment means and variance test for analysis. Statistical analysis was performed using SASS software. Means were compared using Duncan multiple range tests at $P \leq 0.05$. The obtained data were analyzed by Microsoft Excel (2010) program.

RESULTS

The scanning electron micrographs from leaf surface at various stages are shown in Fig. 1. The surface was analyzed before treatment at various stages of growth.

The length of trichomes is presented in Fig. 2. The data shows that the length of trichome in stage I, II and III was increased in 1/3 fc drought (35.7, 34.45 and 39.61 mm respectively).

Fig. 1. Scanning electron micrographs of before (stage I), during (stage II) and after (stage III) flowering. The leaf surfaces were prepared using freeze-drying technique, without treatment.

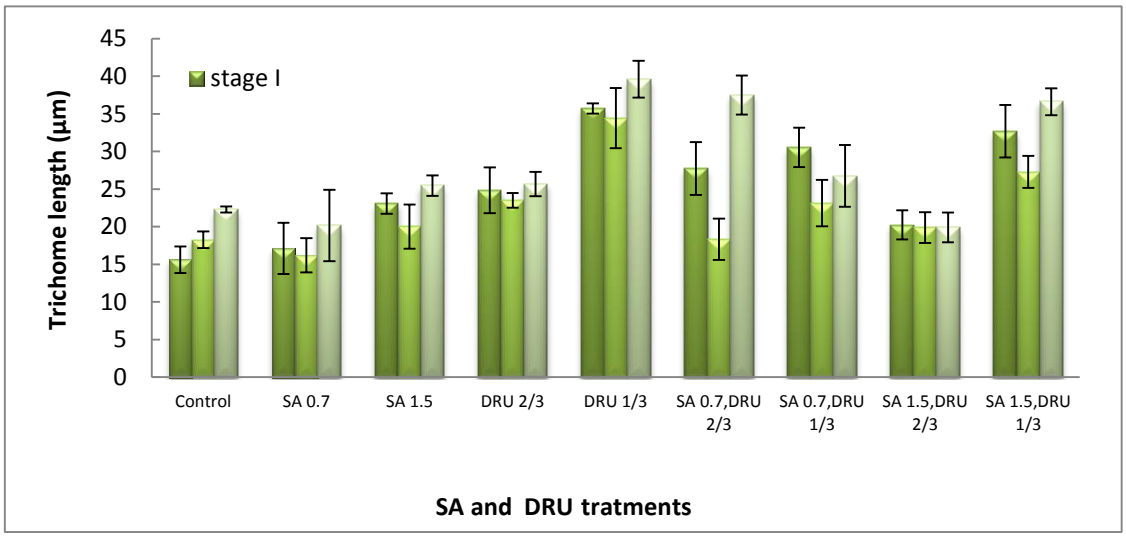

Fig. 2. The effect of salicylic acid (SA) (0, 0.7 and 1.5 mM) treatments, drought stress (DRU) (0, 1/3fc and 2/3fc) and both factors on trichome length of lemon balm at three stages: before (stage I), during (stage II) and after (stage III) flowering.

The concentrations of photosynthetic pigments (chlorophyll a, b, carotenoids and total chlorophyll) are presented in Table 2. Each column belongs to a specific sampling stage. The highest amount of chlorophyll a in the three stages was related to 0.7 mM salicylic acid (7.71, 9.41 and 10.15 mg g^{-1} Fw respectively), that of chlorophyll b in 1.5 mM salicylic acid (6.07, 7.37, and 6.17 mg g^{-1} Fw respectively) and that of carotenoids in 0.7 mM salicylic acid (2.97, 3.27, and 3.59 mg g^{-1} Fw respectively). Total concentration of protein is shown in Fig. 3. It is evident from this figure that protein level was increased at I, II and III stages in 0.7 mM treated salicylic acid and 1/3 fc drought samples (2.59, 5.75 and 7.07mg g^{-1} Fw respectively). The content of MDA determines the extent of lipid peroxidation. Fig. 4 shows its concentration in the treated samples at various stages.

Table 2. Content of pigments in three stages (I, II, III).

	Chl a mg g^{-1}			Chl b mg g^{-1}			Car mg g^{-1}			Chl a+b			Total chl a/chl b		
	I	II	III	I	II	III	I	II	III	I	II	III	I	II	III
Control	7.09ab	8.76ab	9.17abc	5.26a	5.52bc	5.63ab	2.06ab	2.34ab	2.72abc	12.36a	14.29b	14.81a	1.36a	1.62ab	0.13a
SA 0.7	7.71a	9.41a	10.15a	5.42a	6.72ab	5.89ab	2.97a	3.27a	3.59a	13.13a	16.16a	15.99a	1.25a	0.89ab	1.59a
SA 1.5	7.57ab	6.55b	9.81ab	6.07a	7.37a	6.17a	2.95a	3.19ab	3.14ab	13.65a	13.95b	16.04a	1.42a	1.40ab	1.80a
DRU 2/3fc	6.24b	7.35bc	6.85d	1.91b	3.41d	2.88dc	1.51b	1.78ab	1.73c	8.160b	10.75dc	9.74cd	3.26a	2.18a	2.58a
DRU 1/3fc	6.22b	6.65b	6.84d	1.80b	3.34d	2.26d	1.45b	2.08ab	1.83c	8.03b	10.0d	9.10d	3.51a	1.98b	3.41a
SA 0.7, DRU 2/3fc	6.52ab	7.93bc	7.65dc	2.35b	3.58d	3.92abc	2.23ab	2.53ab	2.46bc	8.88b	11.55dc	11.54bc	3.28a	2.30a	1.95a
SA 0.7, DRU 1/3 fc	6.81ab	7.97bc	8.13dc	2.46b	4.26dc	4.42abc	1.58b	1.45ab	1.92c	9.28b	12.23c	12.55b	3.60a	1.90ab	1.85a
SA 1.5, DRU 2/3fc	6.94ab	7.90bc	8.05dce	2.69b	3.31d	4.30abc	1.68ab	2.21ab	2.13bc	9.63b	11.21dc	12.36b	2.61a	2.40a	1.87a
SA 1.5, DRU 1/3fc	6.50ab	8.19bc	8.59bcd	2.58b	3.70d	3.34dc	1.69ab	1.65ab	1.84c	9.08b	11.89c	11.93b	3.48a	2.31a	2.58ba

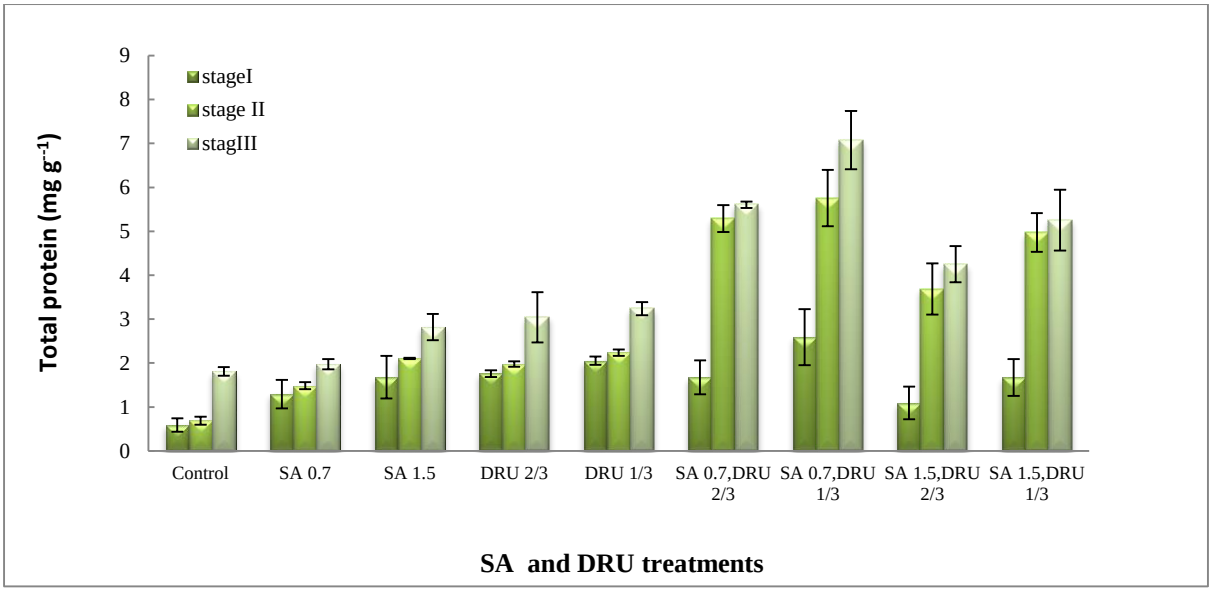

Fig. 3. The effect of salicylic acid (SA) (0, 0.7 and 1.5 mM) treatments, drought stress (DRU) (0, 1/3 fc and 2/3fc) and both factors on total protein content of lemon balm at three stages: before (stage I), during (stage II) and after (stage III) flowering.

It can be observed that the amount of the MDA was increased during first and second stages in 1/3 fc drought treatments (9.70, 10.50 µ mol g-1respectively) and in third stage of 2/3 fc drought treatment (12.12). On the other hand, as shown in Fig. 5, phenol content was increased in stage I of 0.7 mM salicylic acid and 1/3 fc drought (18.00 mg g^{-1} Fw) treated samples as well as in stage II in 1.5 mM salicylic acid and 2/3 fc drought (25.19 mg g^{-1} Fw) ones and then in stage III, in 1.5mM salicylic acid and 1/3 fc drought (25.02 mg g^{-1} Fw). Fig. 6 represents variations in flavonoid concentration at various stages. It was found that the flavonoid content was increased in 0.7 mM salicylic acid and 1/3 fc drought in all steps (0.50, 0.52 and 0.52 mg g^{-1} Fw respectively).

As demonstrated in Fig. 7, the anthocyanin content was increased in the first and third stages in 0.7 mM salicylic acid and 1/3 fc drought (36.95 and 57.93 µmol g^{-1} Fw respectively), as well as in second stage in 1/3 fc drought (40.29 mg g^{-1} Fw).

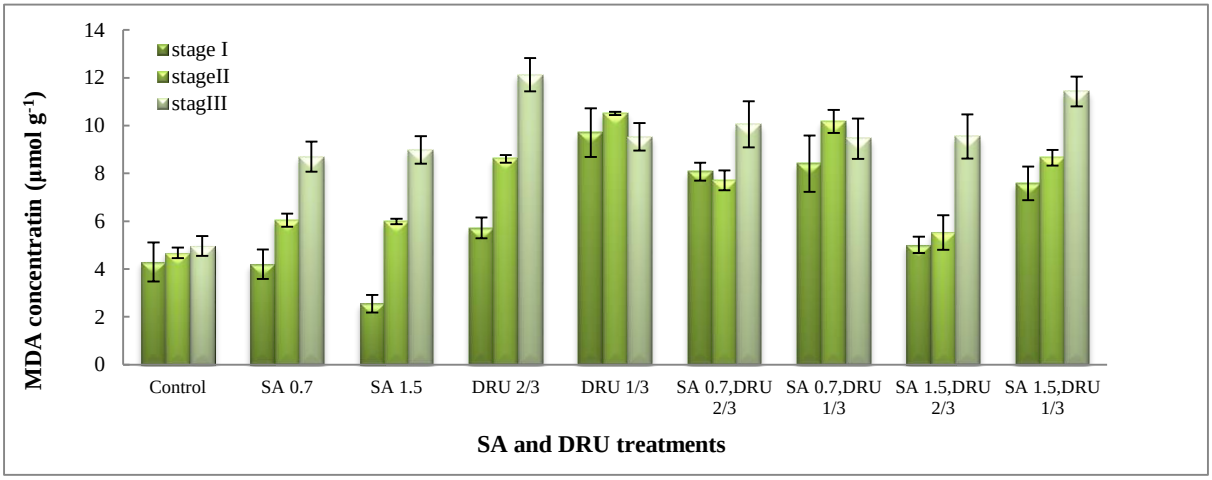

Fig. 4. The effect of salicylic acid (SA) (0, 0.7 and 1.5mM) treatments, drought stress (DRU) (0, 1/3 fc nd 2/3 fc) and both factors on the MDA content of lemon balm at the three stages: before (stage I), during (stage II) and after (stage III) flowering.

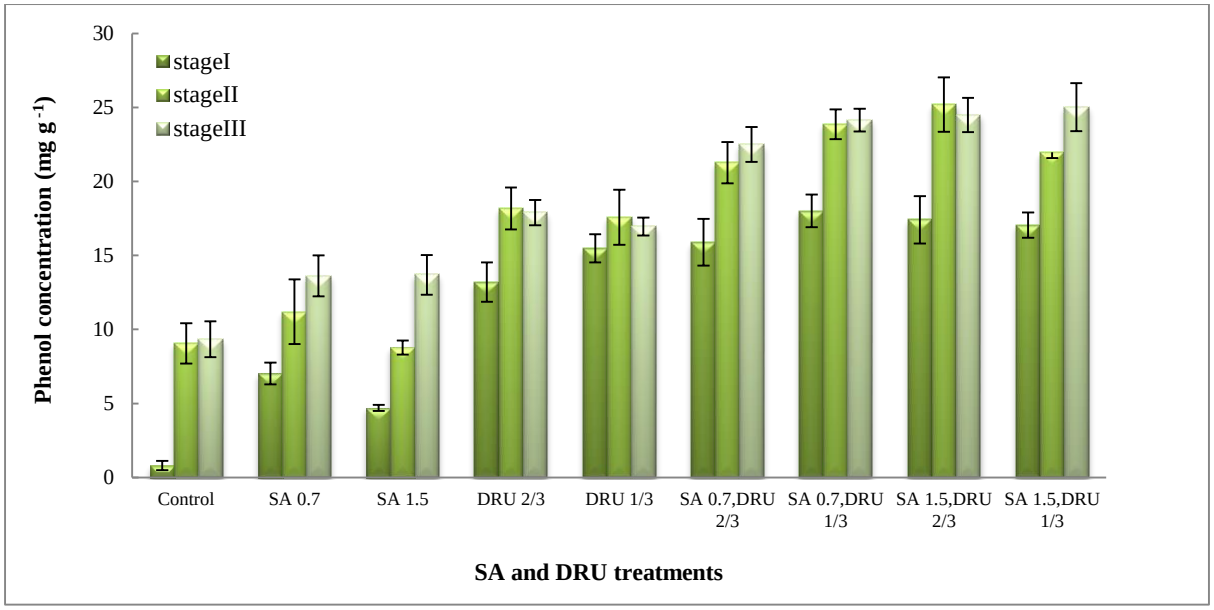

Fig. 5. The effect of salicylic acid (SA) (0, 0.7 and 1.5 mM) treatments, drought stress (DRU) (0, 1/3 fc and 2/3 fc) and both factors on total phenol content of lemon balm at three stages: before (stage I), during (stage II) and after (stage III) flowering.

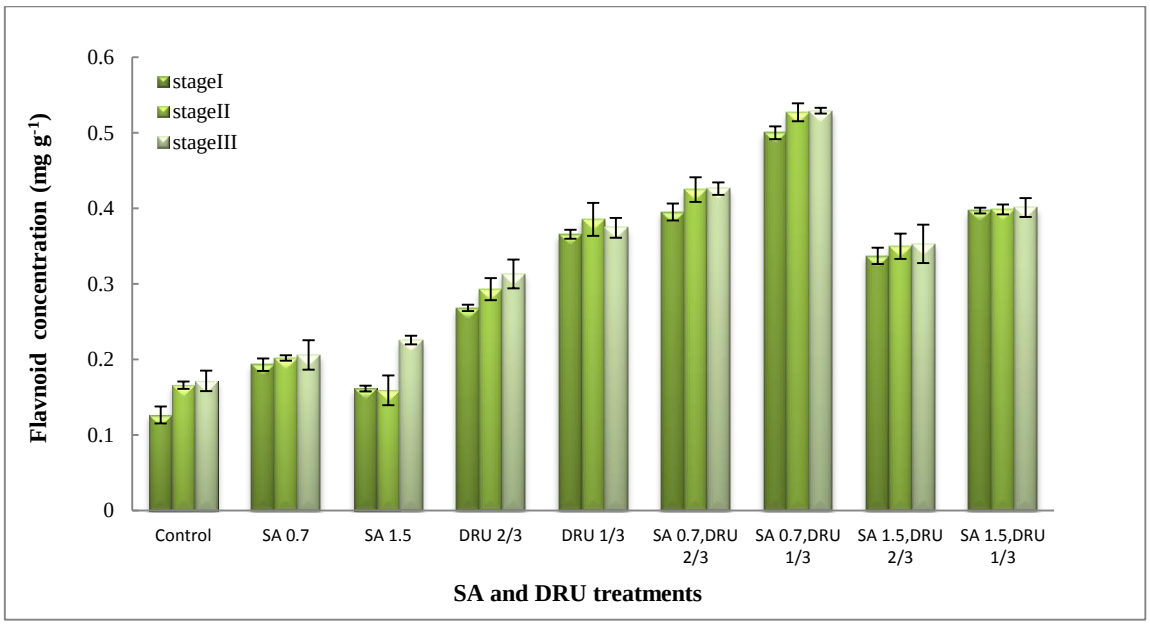

Fig. 6. The effect of salicylic acid (SA) (0, 0.7 and 1.5mM) treatments, drought stress (DRU) (0, 1/3 fc and 2/3 fc) and both factors on total flavonoid content of lemon balm at three stages: before (stage I), during (stage II) and after (stage III) flowering.

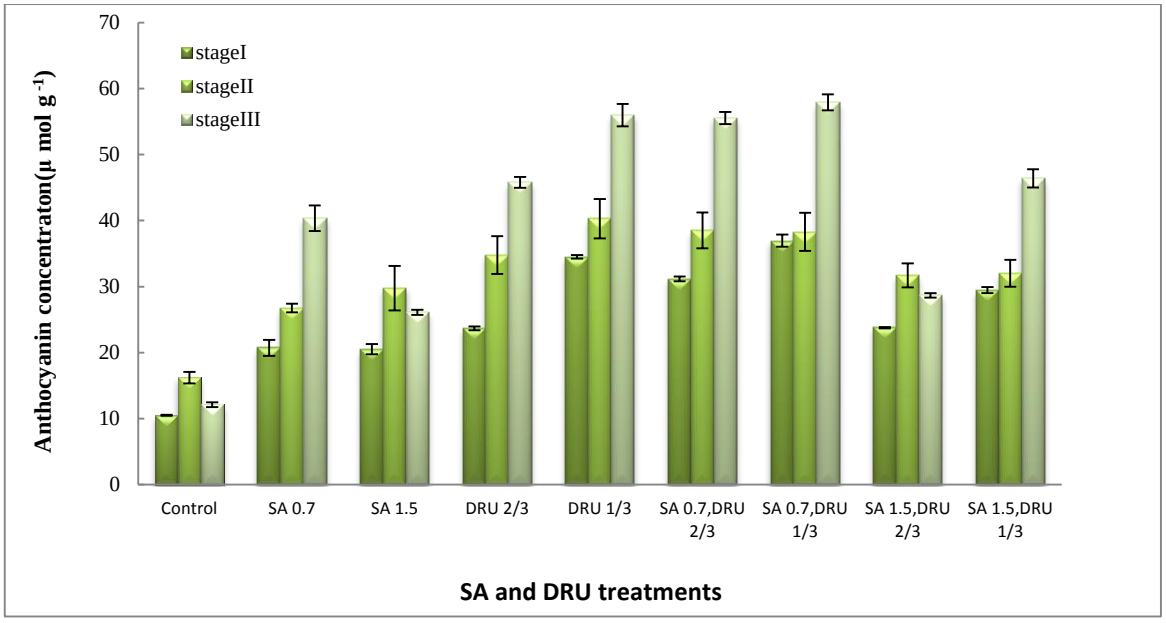

Fig. 7. The effect of salicylic acid (SA) (0, 0.7 and 1.5 mM) treatments, drought stress (DRU) (0, 1/3 fc and 2/3 fc) and both factors on total anthocyanin content of lemon balm at three stages: before (stage I), during (stage II) and after (stage III) flowering.

DISCUSSION

Trichomes

Length of leaf surface trichomes increased significantly under water stress. In contrast, the effect of SA was not significant ($P \leq .05$). Plants have evolved a multitude of defense mechanisms against biotic and abiotic stress factors such as drought, heat, and herbivory (Agrawal *et al.* 2006). Leaf trichomes can protect plants against drought by reducing absorption of solar radiation, which in turn reduces the heat load and minimizes the need for transpirational cooling (Espigares *et al.* 1995).

Photosynthetic pigments

Water stress is a limiting factor for the growth of many plants. A number of studies have revealed that in medicinal plants, drought leads to an increase in secondary metabolites. It has been found that SA can considerably affect the sensitivity of plants to a number of abiotic stresses (Dat *et al.* 1998). In the present study, we were mostly interested to determine the effect of treatment with SA when the plant is subjected to drought stress.

In other words, to determine whether treatment with SA can reduce the possible adverse effects caused by drought stress. As expected, we observed that both drought stress and SA treatment significantly affected photosynthetic pigments (chlorophyll a, a+b and carotenoids). It was found that concentration of the chlorophyll a increased due to treatments with 0.7mM salicylic acid in all three stages, while decreased in both cases of drought 1/3 and 2/3 fc. In support of our results, it has been found that application of 0.7 and 1.5 mM SA led to an increase in production of all pigments. Besides, Ghai *et al.* (2002) have reported that application of salicylic acid to *Brassica napus* improved its chlorophyll content. A more recent research has shown that drought stress could significantly change the amounts of chlorophyll a, b and carotenoids in the plant (Farooq *et al*. 2009). A similar result is reported for cotton subjected to drought stress (Jaleel *et al*. 2008). They found that the ratio of chlorophyll a to b was reduced due to drought stress.

They suggested that decrease in photosynthetic pigments is due to instability of protein complexes and destruction of chlorophyll due

to the increased activity of the chlorophyll degrading enzyme, chlorophyllase.

The result of Jaleel *et al* (2008) indicates that under stress conditions photosystem II protects the plant against low water stress. It is worth indicating that photosynthetic pigments are important for the plant growth and development, mainly for harvesting the light and forming the important reducing powers (Jaleel *et al.* 2008). Furthermore, in support of our results, it has been reported that SA treatments increased chlorophyll a and b as well as carotenoids (Khayatnezhad *et al.* 2011). In contrast, Alaei (2011) reported that under drought stress, the amount of leaf chlorophyll was increased in all wheat genotypes (Alaei 2011). Moreover, reduction in chlorophyll a/b under drought stress has again been reported in some other studies on sunflower and wheat plant (Pastori & Trippi 1993; Sgherri *et al.* 1993). The most important index for photosynthesis is the total content of chlorophyll (Jiang *et al.* 2001). In the case of plants subjected to drought stress, the activity of the Photosystem II (PSII) and Ribulose-1,5-bisphosphate. Carboxylase/oxygenase (RubisCO) decreases, ATP synthesis is stopped and free oxygen is accumulated in chloroplasts (Lawlor *et al.* 2002). The use of salicylic acid could increase the efficiency of PSII and also RubisCO concentration, leading to higher production of ATP for the stabilization of CO_2 (Khan *et al.* 2003).

Protein content

The results obtained from the present study showed that, under drought stress, the protein concentration was significantly increased in samples treated with salicylic acid, compared to the untreated control samples.

This finding could be best understood considering the explanation given by Sakhabutdinova *et al.* (2003). They established that treatment of wheat plants with 0.05 mM SA increased cell division within the apical meristem of seedlings roots which caused an increase in plant growth. It is emphasized that previous studies by other researchers have indicated an induction in expression of genes of proteins that cause resistance (Gapińska *et al.* 2008).

It has been suggested that protein accumulation in all treatments of *Melissa officinalis*, is responsible for protection of plants during considerable water loss (Borovskii *et al.* 2002).

It has also been reported that the content of proteins in plant leaves increases during severe drought (Jiang & Huang 2002).

However, SA could affect the defensive proteins including some types of kinases and RubisCO (Popova *et al.* 1997).

MDA concentration

It was observed that MDA content of the plant leaves subjected to water stress was reduced in response to SA treatment (0.7, 1.5 mM).

By monitoring the content of MDA to show the damage caused by drought stress, investigators have observed an enhanced level of free radicals in plants (Rodrıguez-Rosales *et al.* 1999).

They explained that, osmotic stress causes alterations in membrane lipid composition and properties at a cellular level.

It was further postulated by these researchers that low level of the induced leakiness of membrane is caused by lipid peroxidation resulting from uncontrolled Reactive oxygen species (ROS) increase (Rodrıguez-Rosales *et al.* 1999). Similarly, and in support of our results, a group of researchers have found a significant decrease in the concentration of MDA for drought stressed plants in response to SA pretreatment (Yazdanpanah *et al.* 2011).

It has been reported that when lettuce plant undergoing water stress is treated with SA, its sensitivity of oxidation is decreased leading to lower production of reactive oxygen species. Besides, the amount of MDA is also reduced, compared to control plants (Sayyari *et al.* 2013).

It can be explained that, in addition to improving resistance to drought stress, SA plays a significant role in plant resistance to oxidative stress.

This is also supported by the findings from a team of researchers who mentioned SA

treatment of wheat leaves under water stress conditions resulted in lower accumulation of MDA (Agarwal *et al.* 2005).
Therefore, the lipid peroxidation caused by drought stress was ameliorated by SA treatments.

Total phenol concentration
Treatment with different concentrations of salicylic acid (0.7, 1.5 mM) could be effective on phenol density at different growth stages. Phenolic compounds are secondary metabolites in plants and are known to have important antioxidant activity (Tepe *et al.* 2006). Considering the important role of phenols as antioxidants, it is worth remembering that reduced levels of phenolic compounds in plants could lower its important antioxidant activity.
The relatively high total phenolic content of lemon balm samples observed in this study could have a decisive impact on its antioxidant activity. It could be suggested that increased phenolic content is probably the response of plant's defense system to adjust the tension through enzymatic antioxidant reactions.

Flavonoid content
The results of the present study showed that treatment with 0.7 mM SA at 1/3 fc drought stress significantly increased flavonoid at various growth stages, compared to the control sample.
The special chemical structure of flavonoids suggests that their ability to act as an antioxidant activity is influenced by specific position and structure of hydroxyl groups on the phenol ring (Sharififar *et al.* 2009).
It should be emphasized that flavonoid levels in plants are significantly affected by plant growth regulators (Klessig & Malamy 1994). Researchers have found that in *Taraxacum officinale,* the amount of some flavonoid compounds was significantly increased due to the result of treatment with SA as a growth regulator (Singleton *et al.* 1999).
It is reported that SA is able to highly alter some metabolic pathways needed to synthesize important secondary metabolites (Kim *et al.* 2009).

Anthocyanin content
According to our results, total amount of anthocyanins was significantly increased in the third stage of growth. Anthocyanins are protective pigments that act as free radical receptors and protect plants from oxidative stress (Lin-Wang *et al.* 2010).
Increasing anthocyanins in stress conditions is due to their optical protection role in direct removal of ROS during oxidative stress (Zhang *et al*. 2010).
The present study showed that with an increase in low water stress, the anthocyanin level increases in the plant. In another study, the pigment was shown to be resistant to environmental stresses such as dryness and low heat, protecting the plant against free radicals (Watkinson *et al.* 2006). Also, application of SA with high concentrations has increased the anthocyanin level in *Arabidopsis* (Shan *et al.* 2010).

Conclusions
Based on the results obtained from the present study, the salicylic acid and drought stress affect many physiological and biochemical factors in lemon balm. However, its application with concentration of 0.7 mM, was highly effective on improving the antioxidant properties of lemon balm plant subjected to water stress.
Besides, increasing its concentration to, for example 1.5 mM was even more effective, compared to the control plant.
In conclusion, the use of SA not only reduces the adverse effect of low water stress, but also increases the plant resistance to membrane destruction leading to decreased MDA content. It also enhances the plant growth and development by increasing important secondary metabolites such as carotenoids.

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اثر استرس کم آبی روی متابولیت های ثانویه *Melissa officinalis* L.: نقش اگزوژن سالیسیلیک اسید

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چکیده

گیاه بادرنجبویه در طبقه‌بندی کرونکوئست در تیره نعناعیان، جنس *Melissa* و گونه *M. officinalis* قرار دارد. قسمت‌های مورد استفاده بادرنجبویه ،برگ و سرشاخه‌های جوان و اسانس حاصل از قسمتهای مذکور است. این گیاه به دلیل دارا بودن ترکیبات معطر خاص موجود در اسانس آن، در صنایع دارویی، بهداشتی و غذایی کاربرد فراوان دارد. علاوه بر این، بادرنجبویه، دارای خاصیت آنتی اکسیدانی بوده و حاوی آلفا توکوفرول است. از طرفی، نظر بر این است که تولید متابولیت‌های ثانویه برای سازگاری گیاه نسبت به عوامل نامساعد و تنش‌های محیط زندگی صورت گرفته و به منزله به کار افتادن یک نوع جریان دفاعی در جهت استمرار تعادل فعالیت‌های حیاتی به حساب می‌آید. تنش خشکی یکی از تنش‌های زنده در گیاهان است که علت اصلی آن افزایش میزان تلفات آب، یا کافی نبودن میزان جذب آب و یا ترکیبی از هر دوعامل است که بر اثر آن میزان تلفات آب ناشی از تعرق بر میزان جذب آن توسط ریشه‌ها پیشی گرفته و میزان تنش افزایش می‌یابد. سالیسیلیک اسید ترکیبی فنولی است که جزء فیتوهورمون‌ها به شمار می‌آید و دارای اثراتی برمتابولیسم و بیوسنتز و همچنین فعالیتهای اکسیداتیو و فعالیت‌های زیست شناختی مانند رشد و نمو، فتوسنتز، تنفس، جذب و انتقال یون‌ها، تغییر فعالیت برخی آنزیم‌های مهم و ساختار کلروپلاست است. این تحقیق در دانشکده علوم دانشگاه گیلان در سال جاری انجام شد. برای این منظور بذر گیاه بادرنجبویه پس از ضد عفونی در گلدان‌های مناسب کشت شد و پس از رشد، در مرحله ۶ برگی تحت تنش خشکی قرار گرفت و همزمان هورمون سالیسیلیک اسید بر روی برگ گیاه اسپری شد. برداشت نمونه در سه مرحله (قبل گلدهی، گلدهی و بعد گلدهی) انجام شد و در هر سه مرحله رنگدانه‌های گیاهی، میزان پروتئین، فنل، فلاونویید و آنتوسیانین مورد سنجش قرار گرفت. نتایج نشان داد که اسپری سالیسیلیک اسید بر روی برگها در مراحل رشد ضمن افزایش رنگدانه‌های فتوسنتزی و ترکیبات آنتی اکسیدانی باعث بهبود اثر خشکی و همچنین، بهبود وضعیت فیزیولوژیک گیاه بادرنجبویه می‌شود.

مؤلف مسئول