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Effect of Drought Stress on the Morphology of Three *Salvia sclarea* Populations

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ABSTRACT

To investigate the effects of drought stress on morphology of Salvia sclarea, this experiment was conducted in 2011 in Alborz Research Station, dependent of Research Institute of Forests and Rangelands, Karaj, Iran. The experiment was conducted in split plot in the form of a randomized complete block design with three replications. The main plots were irrigation (at 30 and 60% of field capacity along with a well watered control). The sub plots were three Salvia sclarea populations (Esfehan; Semnan and Karaj). The results showed that drought stress had significant effect on dry matter yield, lateral root numbers, root length, the longest lateral root, root diameter, root volume, plant height and also leaf length and width. Moreover, population had significant effect on dry matter yield and plant height. Among all the measured samples, Karaj population had the highest dry matter yield and plant height; however, Isfahan population had the lowest value of the mentioned traits. Meanwhile, the control produced the highest dry matter yield (724.78 kg/ha), root diameter (10.48 cm), plant height (47.88 cm), leaf length (38.03 cm) and leaf width (19.5 cm). The lowest values of the mentioned traits were achieved in 30% of FC. The longest root (49.21 cm) was observed in 60% of FC and the shortest one (34.11 cm) was recorded in 30% of FC. Karaj population × control produced the highest dry matter yield, number of lateral roots, root diameter, root volume, plant height and leaf length and width. Therefore, Karaj population could be introduced as suitable Salvia sclarea population in Karaj region.

Keywords: clary sage, drought stress, field capacity, medicinal plant, *Salvia sclarea*.

INTRODUCTION

Clary sage (*Salvia sclarea*) is a herbaceous plant of Lamiaceae family which has 58 annual or biennial species in Iran, 17 of them are endemic to Iran [26]. The origin of the plant is reported to be dry sandy soils of Caucasus, Iran and European Coast of Meditranian Sea [1, 21]. For centuries, clary sage has been under attention and has been used the first time to cure the symptoms of insects bite and as a panacea. Moreover, it has been used to improve general body health and for longer life. Nowadays, essential oil of clary sage is used to aromatize tuna, sausages and chicken meet. In addition, it is used as the basic essential oil in perfume industries, to be mixed with other essential oils [1, 5, 6, 12, 22].

Stress is a factor outside plant's body which damages plant growth [13]. Among the abiotic stresses, drought is the most important one which affects plants periodically in some growth stages, or permanently in all life cycle [19].

Drought stress usually occurs when available water in soil reduces and atmospheric conditions increase water loss through evapotranspiration [7]. A primary symptom of low available water to plants is the loss of turgor pressure and reduction of cell development especially in stems and leaves. Reduction of cell development makes the plant smaller in size, which is the characteristic of drought stressed plants. Moreover, drought stress disturbs nutrient absorption and reduces leaves growth. Lower leaf area means lower light absorption and photosynthesis. All these events finally decrease plant growth and yield [25].

Drought stress in induced when moisture at the rhizosphere falls below the permanent wilting point (PWP). So the plant is not able to take up sufficient water, resulting in cell dehydration. Dehydration is reversible until a certain point (elastic point); however, is irreversible if the water loss is too server (plastic point) [3]. However, the time, duration and frequency of drought stress incident, soil properties and so many other factors affect plant tolerance to drought, and different genotypes may also respond differently [10]. Drought stress induces some morpho-physiological responses in plant such as the reduction of leaf area, shoot growth, enhancement of root growth, stomata closure, reduction of growth rate, sudden antioxidants and soluble compounds accumulation, and activation of some enzymes [24]. Safikhani [9] studied the effect of 100%, 60% and 40% FC drought stresses on *Dracocephalum moldavica* and concluded that irrigation at 40% FC (severe drought stress) decreased plant height, leaf area, internodes length, shoot yield and essential oil yield compared with the two other treatments. Stephanie et al. [8] reported that drought stress reduced stem length and root length of *Salvia splendens*. Lebaschi and Sharifi Ashoorabadi [15] concluded that higher drought stress levels reduced plant high and shoot weight in some medicinal plants such as *Salvia officinalis* and *Achillea millefolium*. Sangwan et al. [17] reported that mild drought stress decreased lemon grass height, leaf area and leaf weight. Finally, Ardakani et al. [16] reported that drought stress affected shoot yield, essential oil percentage and yield, leaf yield, stem yield, height, the number of tillers, leaf area, stem diameter and the length of internodes in balm (*Melissa officinalis*).

The objective of this experiment was to assess the response of three *Salvia sclarea* populations to different levels of drought stress.

MATERIALS AND METHODS

This experiment was conducted under field conditions in 2011 at Alborz Research Station, dependent of Research Institute of Forests and Rangelands (RIFR), Karaj, Iran. Alborz Research station is located in 5 km south east of Karaj (35° 48' N, 51° E, 1320 m above the sea level). Average annual precipitation at the site is 235 mm, minimum air temperature is -20°C and maximum air temperature is 38°C. The dominant winds at the area blow from east and south east. The properties of soil at the test site are listed in Table 1.

Table 1. Physic-chemical properties of the test site soil

Depth (cm)	Texture	Sand (%)	Silt (%)	Clay (%)	K (mg/kg)	P (mg/kg)	Na (mg/kg)	C (%)	N (%)	CaO (%)	EC (ds/m)	pH
0-15	L	45	30	25	197.6	10.2	38.7	0.57	0.04	3.1	0.22	8.5
15-30	Sa.C.L	53	26	21	178.6	8.7	32.2	0.68	0.04	3.6	0.19	8.4

The experiment was conducted in split plot in the form of a randomized complete block design with three replications. Each replication consisted of nine treatments. Each plot contained five rows of 50 cm × 3 m. 3 m was left uncultivated between plots and replications. The main plots were irrigation (at 30% and 60% of the field capacity, and normal irrigation), and the sub plots were populations of *Salvia sclarea* (collected from Esfehan, Semnan and Karaj). Seeds were obtained from the Iranian Research Institute of Forests and Rangeland.

Seeds were planted in May 2011, and irrigated immediately. Tinning was conducted after emergence, when plants were at four leaves stage. Irrigation was regularly conducted according to the prepared map. Due to induce the drought stress, 100% of FC was considered as the well water and 60% and 30% of FC were considered as the drought stress treatments. When plants matured enough, at early October when weather was becoming cold, traits such as dry matter yield, the number of lateral roots, branches length, the length of the longest lateral root, root diameter, root volume, plant height, leaf width and leaf length were measured. Finally, data were analyzed using SAS and means were compared using MSTATC.

RESULTS

Dry matter yield. Analysis of variance indicated the significant effect of drought stress and population on dry matter yield; however, the effect of their interactions was not significant (Table 2). Mean comparison of the effect of drought stress on dry matter yield indicated that control and 60% FC had 16.11 and 7% higher dry matter yield, respectively, compared with 30% FC (Table 3). Among the populations, dry matter was higher in Karaj (712.48 kg/ha) and Semnan (696.45 kg/ha) and the lowest in Esfehan (610.36 kg/ha). The population from Karaj and Semnan had 16.73 and 14.1% higher dry matter yield, respectively, compared with Esfehan.

Table 2. Analysis of variance of the effect of drought stress and population on the measured traits

SOV	df	Mean Squares (MS)								
		Total yield	Number of lateral roots	Root length	Length of the longest lateral root	Root diameter	Root volume	Plant height	Leaf length	Leaf width
Block	2	**	**	**	**	**	**	**	ns	ns
Stress (A)	2	**	**	**	**	**	**	**	**	**
Error (A)	4	15551.19	2.66	6.13	0.30	0.0003	59.25	69.25	36.57	8.08
Population (B)	2	**	ns	ns	ns	ns	ns	**	ns	ns
A × B	4	ns	ns	ns	ns	ns	ns	ns	ns	ns
Error	12	2655.26	4.88	7.02	0.44	0.005	44.90	0.27	3.27	2.94
CV (%)	-	7.65	8.22	6.64	2.05	0.91	7.46	1.5	6.4	1.95

ns, non significant; *, significant at P≤0.05; **, significant at P≤0.01.

Table 3. Effect of drought stress, population and their interaction on the measured traits

Treatments	Total yield (kg/ha)	Number of lateral roots	Root length (cm)	Length of the longest lateral root (cm)	Root diameter (cm)	Root volume (cc)	Plant height (cm)	Leaf length (cm)	Leaf width (cm)
30% FC	624.17b	27.33b	34.11b	35.80a	6.11c	1429.44b	22.77c	18.16c	9.61c
60% FC	670.35b	18.00c	49.21a	31.00b	6.54b	1418.33c	34.22b	28.54b	13.83b
Control	724.78a	35.33a	36.30b	30.77b	10.48a	1484.44a	47.88a	38.03a	19.5a
Esfehan	610.36b	26.66a	39.56a	32.5a	7.66b	1445.00a	33.77c	28.08a	13.82a
Semnan	696.45a	26.00a	39.45a	32.33a	7.74a	1442.77a	35.0b	28.27a	15.03a
Karaj	712.48a	28.00a	40.60a	32.88a	7.73ab	1444.44a	36.11a	28.37a	14.08a
S_1P_1	556.38d	28b	33.06c	36.06a	6.03d	1430b	22h	18.1c	9.5d
S_1P_2	643.64bcd	26b	34bc	36a	6.16c	1430b	22.66h	17.83c	9.6cd
S_1P_3	672.48abc	28b	35.26bc	35.33a	6.13cd	1428.33bc	23.66g	18.56c	9.73cd
S_2P_1	591.37cd	17c	49.40a	30.33c	6.5b	1423.33bcd	32.66f	28.8b	12.83bc
S_2P_2	704.37ab	18c	50.13a	30.66c	6.6b	1415d	34.33e	29.06b	15.9b
S_2P_3	715.0ab	19c	48.10a	32b	6.53b	1416.66cd	35.66d	27.76b	12.76bcd
S_3P_1	683.3abc	35a	36.23bc	30.66c	10.46a	1481.66a	46.66c	37.36a	19.13a
S_3P_2	741.3ab	34a	34.23bc	30.33c	10.46a	1483.33a	48b	37.93a	19.6a
S_3P_3	749.67a	37a	38.43b	31.33bc	10.53a	1488.33a	49a	38.8a	19.76a

Means in a column followed by the same letter are not significantly different at P≤0.01.
S_1, 30% FC; S_2, 60% FC; S_3, Control.
P_1, Esfehan; P_2, Semnan; P_3, Karaj.

Plant height. Results indicated that drought stress significantly affected plant height (Table 2). The control and 60% FC resulted in the highest plant height (47.88 and 34.22cm, respectively) and 30% FC gave the lowest plant height (22.77cm) (Table 2). Irrigating the field at 100% FC (control) increased plant height by 39.91% compared with 60% FC and by 110.27% compared with 30% FC. Moreover, 60% FC had 50.28% higher plant height compared with 30% FC. Analysis of variance also indicated the significant effect of population on plant height. Mean comparison indicated that plant height was the highest in Karaj (36.11 cm) and Semnan (35 cm) and was the lowest in Esfehan (33.77 cm). Plant height was 6.92 and 3.17% higher in Karaj compared with Esfehan and Semnan, respectively. Plant height was also 3.64% higher in Semnan compared with Esfehan (Table 3).

The number of lateral roots. Analysis of variance showed that drought stress significantly affected the number of lateral roots (Table 2). However, the effect of population and the interaction of drought stress × population had no significant effect on this trait. Mean comparison indicated that 100 % and 30% FC had the highest number of lateral roots (35.33 and 27.33) and 60% FC had the lowest (18). 100% FC had 96.27% more lateral branches compared with 60% FC (Table 3).

Root length. Results indicated that only drought stress had significant effect on root length; population and the interaction of the two factors had no significant effect (Table 1). Mean comparison represented that 60% FC had the longest roots (49.21cm); however, the control and 30% FC had the shortest roots (36.3 and 34.11cm, respectively) (Table 3). Mean comparison of the interaction of drought stress × population showed that Semnan × 60% FC had the highest (50.13 cm) and Esfehan × 30% FC had the lowest (33.06 cm) root length (Table 3).

The length of the longest internode. According to the analysis of variance, drought stress significantly affected this trait (Table 2). According to the mean comparison, 30 and 60% FC had the longest lateral roots (35 and 31 cm, respectively) and the control had the shortest lateral roots (30 cm) (Table 3). Mean comparison of the interaction of drought stress × population showed that 30% FC × Esfehan had the longest (36.06 cm) and the control × Semnan had the shortest (30.33 cm) lateral roots (Table 3).

Root diameter. Results indicated that only the effect of drought stress was significant on root diameter (Table 2). Mean comparison indicated that root diameter was 71.52% higher in the control compared with the 30% FC. This trait was the highest in the control (10.48 cm) and the lowest in 60% FC (6.54 cm) and 30% FC (6.11cm) (Table 3). Mean comparison of the effect of the two factor's interaction showed that root diameter was the highest in control × Karaj, control × Esfehan and control × Semnan, and was the lowest in 30% × Esfehan and 30% × Semnan (Table 3).

Root volume. Analysis of variance showed that drought stress significantly affected plant root volume (Table 2). Mean comparison showed that the control and 30% FC had the highest root volume (1484.44 cc and 1429.44 cc, respectively), and 60% FC had the lowest root volume (1418.33 cc) (Table 3). Mean comparison of the interaction also showed that the control × Karaj and 60% FC × Semnan had the highest root volume (1488.33 cc and 1415.0 cc, respectively).

Leaf length. Results indicated that drought stress significantly affected leaf length (Table 2). Mean comparison of the effect of drought stress levels on leaf length showed that the control had the highest leaf length (38.03 cm) and 60% FC and 30% FC had the lowest leaf length (28.54 and 18.16 cm, respectively). The control increased leaf length by 33.25% compared with 60% FC, and by 109.41% compared with 30% FC. 60% FC also increased this trait by 57.15% compared with 30% FC (Table 3).

Leaf width. Analysis of variance showed the significant effect of drought stress on leaf width (Table 2). Mean comparison showed that leaf width was the highest in the control (19.5 cm) and in 60% FC (18.83 cm) and the lowest (9.61 cm) in 30% FC. The control increased this trait by 40.99% compared with 60% FC, and by 102.91% compared with 30% FC. Moreover, 60% FC increased leaf width by 43.91% compared with 30% FC (Table 3).

DISCUSSION

The results of dry matter in this experiment are in agreement with these of Carter et al. [18] on alfalfa and Safikhani [9] on *Dracocephalum moldavica*. In drought stressed plants, intracellular spaces and their water content reduces to facilitate water flow to plant tissue; resulting in the reduction of relative water content and yield [14].

Regarding the effect of population on dry matter, it looks that Esfehan region is more temperate than the other two areas and when faced with drought stress, its yield severely reduced. Moreover, seeds from Karaj produced the highest yield because this population have been grown in a climate that made them tolerant to the environmental conditions. Plant height also reduced when drought stress level increased. This is a normal reaction of plants that they reduce their vegetative growth and enter the reproductive stage when faced with drought stress. Lebaschi and Sharifi Asoorabadi [15] studied the effect of different drought stress levels (25, 50, 75 and 100% FC) on some medicinal plants and reported that increasing drought stress level reduced dry weight and plant height in all studied plants. About the effect of drought stress on the number of lateral roots, it was observed that the number of lateral roots was the highest in the control. Studies have proven that when high soil moisture is available to plants, plants do not search soil to find water; reducing their number of lateral roots. These results accompanies with those of Hasani and Omidbaigi [2] on basil and Hoseini and Rezvani Moghaddam [11] on *Plantago psyllium*.

When measuring root length, it was revealed that 60% FC had the longest roots. From this result it can be concluded that root growth is less sensitive than shoot growth to drought stress. So, drought stress will increase root/shoot ratio; increasing plant ability to absorb water and nutrients. The length of the longest lateral roots was the highest in 30% FC. Under dry conditions, plants try to obtain water and nutrients from more distant soil volumes by increasing their root growth. Waterman and Mole [20] in their experiments found that root is less sensitive that shoot to drought stress.

The results of this experiment indicated that root diameter was the highest in the control and the lowest in 30% FC. Misra and Srivastava [4] also found the same results on Japanese mint. Results also showed that drought stress reduced leaf length and width. Leaves are one of the most sensitive organs to drought stress which respond quickly to low available water. In an experiment on two grass lemon species (*Cymbopogon nardus* and *C. pendulus*), drought stress significantly reduce plant height, leaf length, leaf area and leaf weight [17]._Rizopoulou and Diamantoglon [23] reported that drought stress decreased leaf length and increased essential oil content.

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