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Research Article Effect of Soil Texture on *Stachys multicaulis* Benth's Root System Architecture under Drought and Salinity Stress

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Abstract

Background and Objective: Changing the plant's architecture root system in the face of environmental stress is one of the most important mechanisms for adaptation of plants and drought stress and salinity are the most important factors that plants should be resistant against them in term of root morphophysiological changes. Therefore, the aim of this research was an assessment of the root morpho-physiological change of the Iranian medicinal endemic *Stachys multicaulis* under environmental stress. **Materials and Methods:** Drought treatments included 3 day intervals irrigation periods and a set of salinity (0, 5 and 25 ds m⁻¹) were used at a completely randomized design in three different soil textures in pots. Morphological changes of plant's root under stress in three soil texture were studied over the time. **Results:** Drought and salinity had a distinguish effect on plant root properties based on soil texture. Root length and volume increased along with the increasing the severity of drought in three light, medium and heavy soil textures. But root length and volume decreased along with the increasing the severity of salinity (p<0.05). The plant's root length was 24, 10 and 17 cm, respectively for drought, salinity and control sample in heavy soil. Both salinity and drought stress, also had a positive effect on root density (r² = 0.56, 0.30 and 0.68 for light, medium and heavy soil textures, respectively under salinity stress). **Conclusion:** This study indicated that morpho-physiological changes of the *S. multicaulis*'s root under salinity and drought stress is closely dependent on physical properties of the soil.

Key words: Drought, salinity, soil texture, root system, Stachys multicaulis

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Drought stress limits plant growth and development in many regions of the world^{1,2} which is a key determinant for the distribution and productivity of terrestrial vegetation³. Also, along to the water shortage, salinity is a major abiotic stress affecting approximately 7% of the world's total land area^{4,5}. Therefore plants are exposed to a wide and diverse range of abiotic and biotic stress conditions and salt and drought stress imposes a major environmental threat limiting plant growth^{6,7}.

However, plant root is the initial part to be affected by stress^{8,9}. Roots are the very place where plants first encounter drought stress, it is likely that roots may be able to sense and respond to the stress condition¹⁰. Due to the different of environmental stresses and their longevity, soil condition and plant type, different effect may be happen on plant root properties. Plants exposure to low level salinity activates an array of processes leading to an improvement of plant stress tolerance and cope with environmental stresses^{11,12} but higher salinity can have adverse effects on many biological processes and decreases the growth of plant¹³.

In recent years, most research have focused on root architecture and its adaption to drought stress^{12,14,15}. Root morphological and physiological adaption called root architecture refers to the spatial configuration of the root system and root morphology¹⁶.

Root system architecture anchors a plant to the ground, influencing nutrient acquisition from the soil¹⁷, water uptake¹⁸ and plays a major role in plant fitness. Changes in the root architecture are tightly correlated with perturbations in environmental conditions¹¹. However, based on exiting the environmental conditions, plants could adapt their root system architecture and distribution¹¹ and root system adaption contributes a crucial factor to survival for the plant.

Differences in root architecture are of special interest to understand the capacity of plant natural adaptation to a particular environment¹⁹. The root architecture system is depending on water availability and also soil structure²⁰. However, it still know very little of the morphological and physiological change for specific plants species under these two main environmental stresses in different soil texture and this is important means to acquisition of multiple soil resources²¹.

Although, study on morpho-physiological properties change under environmental stress such as drought and salt have been done in some special plant species around the word, but yet didn't shown any research on the effect of the environmental stress on morpho-physiological properties in different soil texture for a special plant species. However there are many different reports about the plant morphophysiological response to different environmental stress in a specific soil texture.

With regard to the above mentioned, present research was conducted to investigate the effect of severe drought and salinity on morpho-physiological properties of the Iranian endemic *Stachys multicaulis* Benth. species root in three different soil texture.

MATERIALS AND METHODS

The research was conducted in an open field in Tiran and Karvan region of the Isfahan province in Iran at spring to summer of 2017. Also laboratory section performed at University of Tehran.

As brief, *Stachys multicaulis* is a wild endemic plant in Iran²². A green bush and has a height of 20-40 cm which covered with simple trichome (Fig. 1). The plant is importance in terms of medicine or other usages.

Soil study of the plant habitat: Filed survey was done for identifying the plant individual stand in rangelands of Iran. Then, soil sampling was taken from the plant's habitat from two different depths (0-15 and 15-50 cm). Physical and chemical properties of soil included pH, EC, OM, N, P, K, sand, silt and clay percentage were measured at soil laboratory of the University of Tehran (Table 1).

Pot culturing: In order to study the effect of salt and drought stress on morpho-physiological properties of the plant root, cylindrical plastic pots were used for cultivation.



Fig. 1: General view of S. multicaulis in the natural habitat

Soil factors		Amount	Soil factors		Amount
Silt (%)	D ₁	32.90±5.42	K (Meq L ⁻¹)	D ₁	82.40±11.13
	D_2	33.10±4.70		D_2	60.50±8.55
Clay (%)	D ₁	34.40±6.22	Caco ₃ (%)	D ₁	12.90±3.25
	D_2	38.60±7.02		D_2	13.80±2.64
pН	D ₁	7.87±0.40	Ca^+ (Meq L ⁻¹)	D ₁	0.80±0.05
	D_2	7.88±0.24		D_2	0.72 ± 0.04
EC (ds m ⁻¹)	D ₁	0.93±0.12	Cl^{-} (Meq L^{-1})	D ₁	0.70±0.02
	D_2	0.90±0.10		D_2	0.64±0.03
SAR	D ₁	0.35±0.03	HcO_{3-} (Meq L ⁻¹)	D ₁	0
	D_2	0.31±0.01		D_2	0
Na (Meq L ⁻¹)	D ₁	0.49±0.02	Co ₃₋₂ (Meq L ⁻¹)	D ₁	3.41±0.22
	D_2	0.33±0.03		D_2	3.02±0.13
OM (%)	D ₁	1.60±0.08	So_{4-2} (Meq L ⁻¹)	D ₁	0.29±0.02
	D_2	1.40±0.12		D_2	0.21 ± 0.01

D₁: 0-15 cm and D₂: 15-50 cm

 Table 2: Summary of used soil properties and soil ingredient for pot culturing

Soil texture	Clay (%)	Silt (%)	Sand (%)	Texture	Bulk density (g cm ⁻³)
Light	5.00±0.00	25.00±0.00	60.00±0.00	Sandy loam	1.52
Medium	15.00±0.00	42.00±0.00	43.00±0.00	Loam	1.43
Heavy	40.00±0.00	30.00±0.00	30.00±0.00	Clay loam	1.35

Preparing soil for planting: Soil samples were taken for pot culturing. Habitat soil was included 24, 32 and 42% clay, silt and sand, respectively. Three different soil textures were prepared on the basis of changes in soil constituents i.e., percentage of sand, silt and clay²³ (Table 2). Therefore three soil textures including light, medium and heavy prepared adding clay and sand manually.

Plant materials: Due to the problem in germination of *S. multicaulis* seeds, plant scion was cultivated in pots. For this purpose, plant scions were gathered from the field (at the beginning of spring 2016) and transfer to the home garden in order to cultivate in pots.

Applying water and salinity stress: A set of salinity (5 levels of NaCl including 0, 5, 10, 15, 20 and 25 ds m⁻¹), drought treatments (5 levels; including 3 day intervals irrigation periods of 3, 6, 9, 12 and 15 days) in 3 different soil texture (light, medium and heavy) and 4 replicates were prepared in a completely randomized design. About 35 days after culturing, treatments of drought and salinity were applied.

Root morpho-physiological measuring: After 60 days, the following plant root morpho-physiological properties were measured: root length, root volume, root moisture, root density and general root formation. Root sampling and biomass measuring were conducted according to the previous study²⁰. Finally, to analyze the collected data, one-way ANOVA and the *post hoc* test (Duncan grouping) were performed using SPSS software at significance level of 5%.

RESULTS

A significant difference was observed among the all measurements of the root properties under salinity and drought stress for different soil textures. The effect of soil texture on root architecture was considerable as well. The most change in root length under drought stress is related to heavy soil texture while in salinity stress, the most change was occurred for light texture (Table 3).

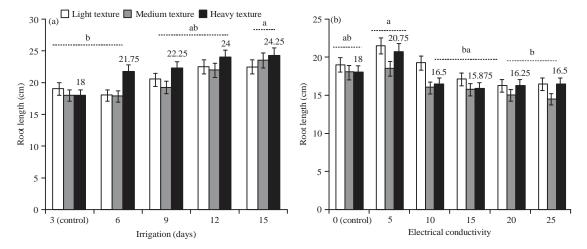
The most change of plant root length, without considering soil texture, occurred in drought stress. The severity of the drought and salinity had various effects on the length of root (Fig. 2).

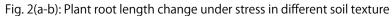
Although some factors such as root length increased by severe stress, but the total volume of the plant root decreased. This factor was nearly identical for three soil texture. The most change for root volume was accrued in pots which were under salinity stress (Fig. 3).

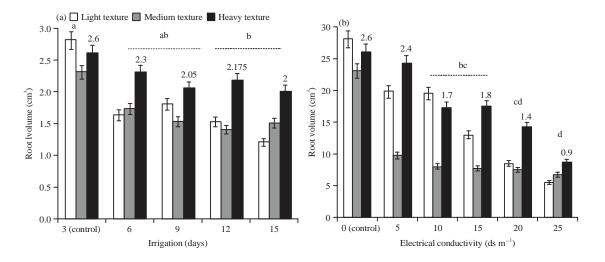
Most change of root dry weight happened under different levels of drought and salinity treatments in heavy soil texture. The least root weight happened for light soil texture had influence on the changes under stress (Table 4).

The results showed a decreased trend of the root moisture content of *S. multicaulis* under different level of drought and salinity treatments. The results also indicated that plant could preserve its moisture at the severe stress in both salinity and drought as a durable plant to environmental stress (Fig. 4). Both environmental drought and salinity stress have similar effect on moisture content of the studied root plant species.

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Soil texture	Variation resources	Drought				Salinity			
		df	SS	MS	Sig.	df	SS	MS	Sig.
Light	Between groups	4	66.000	16.500	0.03*	5	81.802	16.360	0.001**
	Within groups	15	39.000	2.600	-	18	32.687	1.816	-
	Total	19	105.000	-	-	23	114.490	-	-
Medium	Between groups	4	101.000	25.250	0.054*	5	52.208	10.442	0.021*
	Within groups	15	126.937	8.462	-	18	142.750	7.931	-
	Total	19	227.938	-	-	23	194.958		
Heavy	Between groups	4	100.700	25.175	0.001**	5	67.219	13.444	0.027*
	Within groups	15	44.250	2.950	-	18	72.688	4.038	-
	Total	19	144.950	-	-	23	139.906		-

Table 3: Change of root length of plant under drought and salinity in different soil texture

**Significant regression relationship at the level of one, SD: Standard deviation, df: Degrees of freedom, SS: Sum of squares, MS: Mean of squares

Morphological observations of the plant's roots showed that lateral root will be eliminated against the environmental stress. Plant root will be changed to longer root under drought stress but salinity stress can have a negative effect on root length in comparison to control samples (Fig. 5). The density of the plant root was increased along with the increasing severity in drought and salinity stress. The most density was observed in higher level of salinity stress (Fig. 6). Relation between root density and soil texture (Clay content) in different level of salinity stress indicated that clay

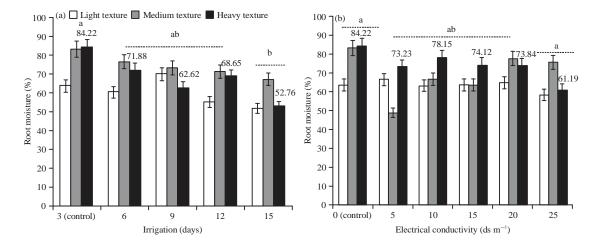


Fig. 4(a-b): S. multicaulis root moisture content change under stress in different soil textures

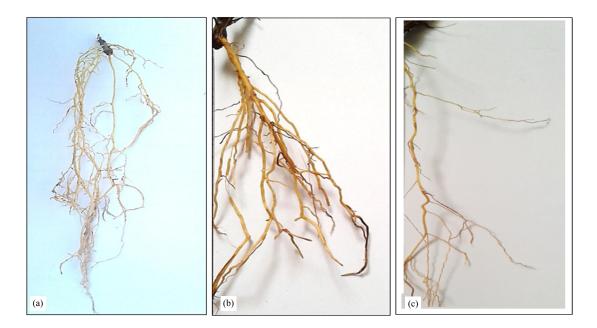


Fig. 5(a-c): Morphological change of the *S. multicaulis* root under stress in heavy soil texture, (a) Control, (b) Salinity (25 ds m⁻¹) and (c) Severe drought

Sig. 0.031* _

0.025* -

0.001** _ _

Soil texture	Variation resources	Droug	nt			Salinity	/	
		df	SS	MS	Sig.	df	SS	MS
Light	Between groups	4	0.330	0.083	0.024*	5	1.184	0.237
	Within groups	15	1.453	0.097	-	18	3.454	0.192
	Total	19	1.783	-	-	23	4.637	-
Medium	Between groups	4	101.000	25.250	0.032*	5	2.477	10.442
	Within groups	15	126.937	8.462	-	18	2.302	7.931
	Total	19	227.938	-	-	23	4.779	
Heavy	Between groups	4	100.700	25.175	0.001**	5	6.659	1.332
	Within groups	15	44.250	2.950	-	18	3.672	0.204
	Total	19	144.950	-	-	23	10.332	

**Significant regression relationship at the level of one, SD: Standard deviation, df: Degrees of freedom, SS: Sum of squares, MS: Mean of squares

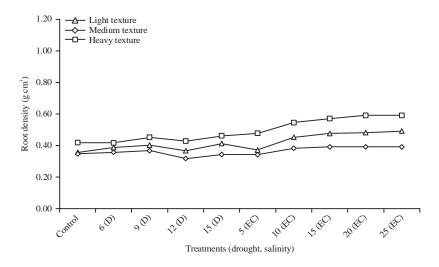


Fig. 6: S. multicaulis root density change under stress in different soil texture

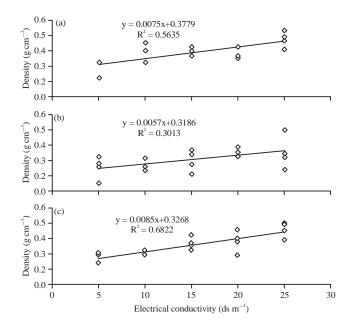


Fig. 7(a-c): The relation of *S. multicaulis* root density and soil texture (Clay content) in different level of salinity stress for different soil texture, (a) Sandy, (b) Loam and (c) Clay

soil (s) has most positive effect on root density. On the other hand, plant in clay soil uptake and store more nutrition and mineral in itself roots (Fig. 7).

Contrary to public opinion, loamy soil has less positive effect on plant properties without stress (Salinity of 0 and drought of 3 day irrigation were as control samples). Generally, all measured plant features, in clay soil showed higher values.

DISCUSSION

Root architecture modification is an important plant response to nutrient availability²⁴ and also osmotic stress caused by drought and soil salinity affect plant root system²⁵. Therefore, the need to develop crops to higher salt tolerance has incased strongly, due to increased salinity problems²⁶ and severe drought as well. In this regards, roots are the primary plant elements which are faced to environmental stress and play an important role in plant tolerance and productivity. Water stress due to drought remains the most significant abiotic factor limiting plant growth and development²⁷ and understanding the mechanisms of drought tolerance and breeding for drought-resistant crop plants has been the major goal of plant biologists and crop breeders¹⁰. Regarding to the above, this is important to know the mechanism of root architecture change under environmental stress and conditions.

The results of the *S. multicaulis* cultivation in different soil texture under drought and salinity stress was noticeable. Generally, drought and salinity had a distinguish effect on some of root properties. Drought increased root length along with the increasing the severity of drought in all three soil texture and the most root length was shown in clay soil. The right architecture in a given environment allows plants to survive periods of water deficit and compete effectively for resources. But in other study, it has been reported that both silty and clay soil reduced the root growth²⁸. The most positive and significant effect (p<0.05) of soil texture on plant factors, was resulted in clay soil (Fig. 2-4). Although some factors such as root length increased by severe stress, but the total volume

of the plant root decreased (Fig. 3). Other study disclosed that drought-tolerant plants tended to distribute relatively more roots in the soil volume²⁹. But in *Cichorium intybus* didn't show a significant different in root length and lateral branches (at p<0.01)³⁰. Also, in other study it was found that disclosed that high NaCl concentrations significantly impeded plant growth resulted in reduction in plant height, root length³¹. Therefore under conditions of drought stress plants root undergo key physiological change that confer greater resistance to low water availability.

Root dry matter (weight) indicated the good or bad soil condition for plants. Salinity decreased directly both dry weights and length and also volume of the plant root in the three different soil textures. But drought stress increased dry weights and length of the plant root and decreased root volume. Similar to this study a negative effect was reported of saline water on the morphological properties of the *Panicum antidotale* root in a soil of loamy sand texture³². Also, in this research root density increased with increasing severity of environmental stress and treatment of salinity in clay soil had the most positive effect on root density (Fig. 7). This factor may change resistance and adaptability of plants in different environments under stress.

The observations of the plant's roots surface also showed that lateral roots are decreased under the environmental stress (Fig. 5). This may be an auto regulation system for plant to resistance against stress. In another study also reported that the stress induced formation of lateral roots the stress induced formation of lateral roots the stress induced formation of lateral roots³³. The relationship between root density and stresses applied to the plant has been shown to be well established. This relationship is much stronger and more specific in the soil with heavy under two salinity and drought stress ($R^2 = 0.3$ -0.68). But in general, the plant density changes under drought stress were more than drought stress. Both severe drought and salt stress causes hairy and fine roots loss in the plant.

CONCLUSION

S. multicaulis showed different root morphophysiological changes against salinity and drought stress. Probably this plant adapts itself to the harsh condition with specific root and aria parts morpho-physiological treats. Drought and salinity had different effect on morphophysiological root properties based on soil texture. This finding will help us for successful establishment of the *S. multicaulis* to the stressful environments. Cultivated plant in medium soil texture had the least performance under salinity, drought condition.

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