



Investigating the effect of drought stress induced by mannitol on germination and morphological properties of wheat inbred lines

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Abstract

To investigate the response of inbred lines of M82 and Babax wheat series to drought stress, which induced by mannitol, on growth stage of plant, a factorial experiment with a completely randomized design with 2 replicates in normal and drought stress condition on 169 in board lines, was performed. In this research the mannitol with -8 Mpa osmotic pressure was used and the morphological properties such as stem and root length, wet weight of stem and root, dried weight of stem and root, proportion of stem length to root length, wet weight of stem to root, seedling length, proportion of dried weight of root to stem, sum of root and stem wet weigh and sum of dried weight of stem and root were studied and statically analyzed. The results indicated that drought stress had significant effect on investigated properties. Results of cluster analysis showed that in drought condition, lines with numbers 11,9,8,1 and 19 in comparison to others were greater in stem length, dried weight of stem, length of seedling and sum of dried weight of root and stem. In drought condition, some of lines individually were great in some studied parameters, such as lines with number 113 and 109 in root length, 8th line in stem and seedling length, number 9 in stem wet weight. The stem parameter can be used as the best property to select the drought tolerance lines.

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Introduction

Hexaploid wheat (*Triticum aestivum* L.) ($2n=6x=42$) has a large genome size of about 17,300 Mb which is approximately 35 times and 110 times larger than that of rice (*Oryza sativa* L.) and Arabidopsis, respectively (Hussain and Rivandi, 2007).

Wheat is the most widely adapted major crop and is grown on a larger land area than any other crop worldwide (Reynolds *et al.* 2011; Munns and Richards, 2007).

Drought in agriculture refers to water deficit in the root zone of plants and results in yield reduction during the crop life cycle (Rampino *et al.*, 2006; Passioura, 2007; Nevo and Chen, 2010; Ji *et al.*, 2010). Therefore, drought tolerance is defined as the ability of plants to survive and reproduce under water deficit conditions (Fleury *et al.*, 2010).

Drought stress is one of the main yield limitations in crop production. Direct and indirect selection is performed for developing the drought tolerant varieties. Though this method is effective but is time consuming. Drought stress reduces the crop yield up to 70 percent (Boyer, 1982). In all over the world drought is the main limitation parameter in crop production which affects the efficiency of farming (Hussain *et al.*, 2011). The simplest definition of drought in agricultural texts is, when the available water for farming is less than needed water for plant growth and developing it would be drought condition (Deikman and *et al.*, 2011). Drought tolerance is the ability of plant to survive in limited water condition (Turner, 1989).

Drought stress is the most important environmental factor which reduces the growth and yield of many crops, especially in arid and semi-arid regions. The most important effect of drought on plant is reduction of photosynthetic material to fill the grains and reduction of grain filling period (Reddy. *et al.*, 2004). Over all, with considering the proportion of arid areas in Iran to the world, Iran is located on the drought belt land. 61% of Iran's area is covered with arid and

very-arid climate which is 3.1 times more than the global rate.

Totally, Iran allocates 1.2% of world lands, dedicates 2.4% of desertification phenomena without any vegetation and has placed 3.08% of world's deserts in it.

In developing countries which dedicate 37% of wheat growing areas in the world, drought is the major factor in decreasing wheat yield (Rajaram, 2001). Along with rice and corn, wheat is one of the most important cereal crops in the world, which produces more than 60% of calories and proteins in human food (Mohammad Ali Jahanbin and *et al.*, 2012). Water is the most essential ingredient of plant structure and can vary from 80 to 90% of plant weight depending on plant species and tissues type (Maybodi, *m, et al.*, 1381).

Usually to create an artificial bed with controlling water potential, the solid material with high molecular weight is used which doesn't have any role in tissues nutrition and is not absorbed by plant. Mannitol a material with high molecular weight by creating a solution with same condition as normal is used in most drought tolerance experiments.

So, this investigation carry out for the study of drought stress induced by mannitol on germination and morphological properties of wheat inbred lines

Materials and Methods

The studied population consisted of 167 new generated inbred lines of F8 derived from across between two varieties of spring hexaploid and semi-dwarf wheat called Seri M82 (moderately susceptible to drought and environmental stresses) with Pedigree (MX 196-97M 31BWSNS- 1) and Babax (tolerance to drought and environmental stresses) with Pedigree (CM92066-J-OY-OM-OY-4M-OY-OMEX-48BBB-OY) in physiology laboratory of research center for agriculture natural resources of Yazd as a factorial design with 167 inbred lines of M82 and Babax series of wheat parents in a completely randomized

design with two replicates and in normal and drought condition in 1393 was performed. For this purpose 169 in boards of Babax series of wheat were placed in water containing mannitol with -0.8 Mpa potential. 100 clean and antiseptic seeds were placed in each petri dishes which first two petri dishes were in normal condition (containing distilled water) and the other two petri dishes were allocated in drought condition (mannitol solution). The temperature and humidity of germinator device was considered on 25 c and 75% respectively. On the first day 10 cc distilled water or mannitol solution were added to every petri dishes separately and in the other days, 5cc of solutions were added and went it on up to 7 days. In 8th day 5 bushes were randomly collected from each petri dishes and length of root and stem, wet weight of stem and root were measured. After drying the collected bushes in room temperature in 24 hours,

dried weight of root and stem were measured. Then the proportions of wet and dried weight of root, length of seedling and sum of wet and dried weight of plant calculated. Excel, SAS and STAT GRAPH software were used to perform calculations. The aim of this study was investigating the tolerance of new generated inboard lines to drought stress in different seedling conditions in laboratory based on length of stem, root and their wet and dried weights.

Results and discussion

This research indicated that drought stress in lines causes decline in number of roots, length of stem and increase in length of root. Descriptive statistics such as mean, minimum, maximum, range, standard deviation and coefficient variation for each of these characters represent diversity in some of studied parameters (table 1).

Table 1. Descriptive statistics for traits inbred wheat genotypes under drought stress.

Traits	Mean	StdDev	Min	Max	Range	CV(%)
Shoot length	2.01	0.541	0.55	4.93	4.38	4.30
Root length	4.31	0.826	1.96	6.53	4.57	2.78
Shoot wet weight	12.45	3.68	4.35	37.95	33.6	3.76
Root wet weight	11.47	4.22	6.00	33.55	27.55	5.78
Shoot dry weight	4.24	0.912	1.58	8.11	6.53	3.51
root dry weight	5.57	1.09	2.23	8.78	6.55	3.18
root length/shoot length	2.25	0.47	11.18	4.38	3.20	3.57
Shoot wet weight/ root wet weight	0.97	0.32	30.48	2.25	1.77	6.14
shoot dry weight/root dry weigh	1.37	0.33	30.63	2.74	2.10	4.29
Plant length	6.31	1.25	2.60	11.03	8.43	3.79
Sum weight wet shoot and root	23.92	6.69	11.97	54.18	42.22	5.18
Sum weight dry shoot and root	9.82	1.63	4.00	14.28	10.28	2.94

Since various cultivars with various traits of great variation, Verdict based on one or more of the correct adjective morphologically doesn't seem, therefore, to select the best genotypes in addition to the use of the method of calculating the correlation of different statistical methods, benefited from that one of these methods is the cluster analysis. This analysis was performed by Ward method and based on square of Oghlidos distance as a similarity criterion. Results showed that the clusters were significant.

tolerant 40 lines of winter wheat cultivars examined and The results were that Tolerant cultivars in terms of the length, wet weight and dry weight of root and shoot and root to shoot ratio higher have and The results were that of this ratio can be used to evaluate drought tolerance and higher ratio indicates a higher drought tolerance.

Under drought stress condition 9 groups were formed (table 2). In cluster groups measuring mean comparisons was based on the parameters and group 1 had the most stem length, root wet weight, dried

(Nabi Zadeh *et al.*, .1386) The conditions of drought

Tabatabai *et al.*

stem weight, seedling length and sum of stem and root dried weight. In drought condition, lines with numbers 11,9,8,1 and 19 in comparison to others were greater in stem length, dried weight of stem, length of seedling and sum of dried weight of root and stem.

Researchers have reported that reducing the growing shoot and root growth increases plantlets desert. Because of transpiration decrease and root penetration into the depths soil for achieve deep water resources and subsequently increase survival plantlet (Zeng *et al.*, 2010).

Table 2. Number genotypes subjected to drought stress cluster.

Cluster	Number	Line superior
1	18	2-8-12-17
2	33	10-111-112-116-130-129
3	16	15-24-27-34-40-36-38-39-59-43-44
4	27	20-50-52-56-62-66-81-84-88-91-159-165
5	4	22-33
6	27	28-109-143-144-145-146-148-153-154-160
7	1	***
8	23	46-48-51-54-74-76-80-90
9	20	47-55-71-83-83-87-89-97

Table 4. Eigenvalue, Proportion and Cumulative variance of extract factors in stress drought

Factors	Eigenvalue	Percent of Variance	Cumulative Percentage
1	5.43251	45.271	45.271
2	2.6842	22.368	67.639
3	1.84862	15.405	83.044

Factor analysis was performed using principal components method and varimax rotations on data, 3 main components were selected based on the high values of components, these explained 83% of total data variations. First factor indicated 45.27% of data

variation. Some of studied parameters such as stem length, seedling length and sum of dried weight of stem and root had large positive loading which plays the most important role in first factor explanatory and describes the stem characteristics of plant.

Table 5. Factor analysis for under studied traits in stress condition.

Traits	Factor1	Factor 2	Factor 3
shoot length	<u>0.368499</u>	-0.0950583	-0.113119
Plant length	<u>0.353838</u>	0.0413123	-0.355879
Sum weight wet shoot and root	0.322815	0.12323	<u>0.424676</u>
Sum weight dry shoot and root	<u>0.377347</u>	0.107261	-0.0853885
Root length	0.294311	0.124736	-0.464558
Shoot wet weight	0.307216	-0.135102	0.274712
root wet weight	0.244153	0.313264	<u>0.43406</u>
Shoot dry weight	0.33727	-0.235531	0.0580768
root dry weight	0.283713	<u>0.35804</u>	-0.17669
root length /shoot length	-0.206196	0.302466	-0.25916
Shoot wet weight / root wet weight	-0.0871918	<u>0.507809</u>	0.277429
Shoot dry weight / root dry weight	-0.0862163	<u>0.545526</u>	-0.120419

Second factor accounts for 67.63% of data variations. Root dried weight, wet weight of root to stem and dried weight of root to stem had large positive loading

which had the most important role in explanation of second factor and dedicates to plants biological properties.

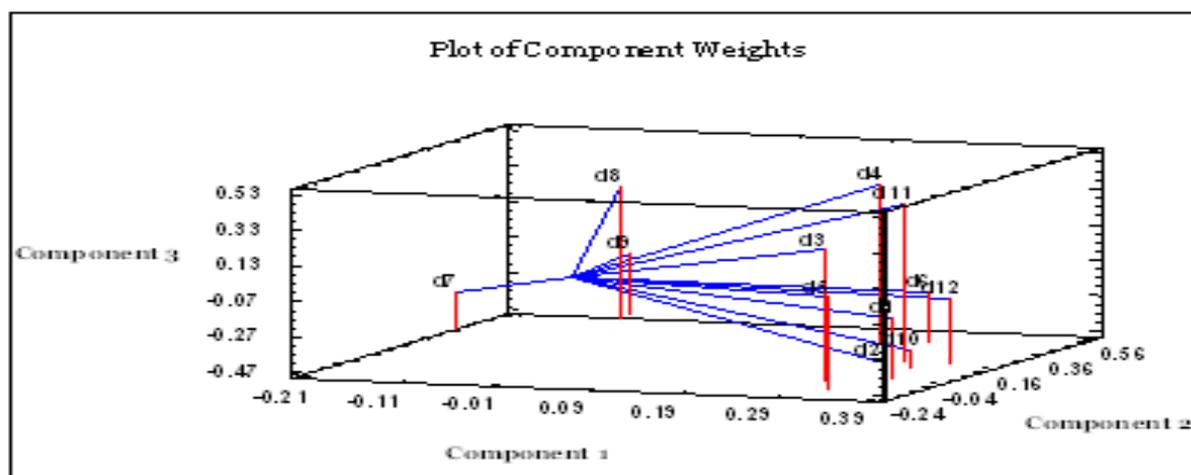


Fig. 1. d1= shoot length d2= Root length d3= Shoot wet weight d4= Root wet weight d5= Shoot dry weight d6= root dry weight d7= root length/shoot length d8= shoot wet weight/root wet weigh d9= shoot dry weight/root dry weigh d10= Plant length d11= Sum weight wet shoot and root d12= Sum weight dry shoot and root.

Third factor applies for 83% of data variations. Properties such as root wet weight and sum of wet weight of root and stem had great positive loading and the most important role in third factor explanation. This factor characterizes ground organs of plant. In general according to the results of factor analysis, it is concluded that since a property's controlling genes in a factor can affect the other significant properties of same factor, for repairing each of the factors, associated properties to the factor must be considered. In other words, a controlling factor or gene can be a common factor. So the results of this section can be used in plant repairing, particularly in indirect selection. In conclusion for repairing wheat in drought stress, more attention must be considered to stem length, wet and dried weight of stem to root proportion and plant length.

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