IDENTIFICATION OF WHEAT GENOTYPES UNDER PEG INDUCED DROUGHT STRESS ON SEED GERMINATION AND SEEDLING GROWTH.

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Abstract

Wheat is a significant cereal crop that provides various food products with high nutritional value. Pakistan is one of the countries that suffers from severe water scarcity and it is worsening over time. For development of water deficit tolerance genotypes, it is crucial to assess the available germplasm for water deficit tolerance. A study was conducted to evaluate commercial wheat cultivars against osmotic stress. Eight wheat genotypes were evaluated under 0 MPa and -0.6 MPa by using PEG 6000 to induce osmotic stress at seedling stage. The results indicated that wheat cultivar Bhakkar star had high percentage of germination under water deficit conditions, root shoot ratio near 1 and maintained relative water contents followed by Markaz-19.

Keywords: Wheat, Water deficit stress, Physio-morphic attributes, Assessment

Introduction

Wheat is one of the most important cereal crops in the world, providing food and feed for humans and animals, as well as raw material for various industries. Wheat is grown in almost every continent and under a wide range of climatic and soil conditions. According to the Food and Agriculture Organization of the United Nations (FAO), global wheat production in 2022/23 is estimated at 783.3 million tonnes, slightly higher than the previous year's record of 779.3 million tonnes. Wheat is the second most produced cereal after maize, accounting for about 30 percent of the world's total cereal output.

Wheat is the main staple food crop in Pakistan and accounts for about 40 percent of the total cropped area and 36 percent of the value added in agriculture. Wheat is an important source of income and food security for smallholder farmers, who produce about 80 percent of the total output. Wheat is also a key ingredient for livestock feed, especially for the poultry sector, which consumes about 10 percent of the total wheat supply. Wheat output is important for food security, poverty alleviation, and economic stability in Pakistan. It also provides about 60 percent of the daily caloric intake and 42 percent of the protein intake of the average Pakistani.

Wheat production is affected by various factors, such as weather, pests, diseases, input availability, and market prices. Among these, climate change is considered one of the most serious challenges for wheat production in the future (Zhao *et al*., 2015). Climate change can alter the temperature, rainfall, and CO_2 levels, which can have both positive and negative impacts on wheat growth and yield. For instance, higher temperatures can reduce the length of the growing season and increase water stress, while higher CO2 levels can enhance photosynthesis and water use efficiency. Climate change can also increase the frequency and intensity of extreme weather events, such as droughts, floods, heat waves, and frost, which can damage wheat crops and reduce yields. Moreover, climate change can affect the distribution and severity of pests and diseases, such as wheat rusts, aphids, and nematodes.

According to a recent study by FAO, global wheat production could decline by 6 percent by 2050 due to climate change. The study also projected that wheat production in Asia could decrease by 8 percent by 2050, with South Asia being the most affected sub region. The study suggested that adaptation measures are needed to cope with the impacts of climate change on wheat production, such as developing heat- and drought-tolerant varieties, improving irrigation and soil management practices, enhancing pest and disease control, and diversifying cropping systems.

Wheat is a significant cereal crop that provides various food products with high nutritional value (Ginkel and Ortiz, 2018). As the world population and the regions where wheat is a staple food grow, so does the demand for this crop (Murgan and Kannan, 2017). Pakistan is one of the countries that suffers from severe water scarcity and it is worsening over time (Anonymous, 2023). Therefore, it is crucial to assess the available germplasm for water deficit tolerance (Murgan and Kannan, 2017). This assessment will help in recommending wheat cultivars for areas with limited water resources. Some morphological and physiological traits have been identified as useful indicators for differentiating genotypes under water stress conditions (Gornicki and Faris, 2014). Based on this, a study was designed to screen a wheat genotypes under water scarcity conditions and evaluate the performance of different genotypes using morphological and physiological parameters. The experiment was conducted under controlled conditions in peteri dishes with use of PEG.

Materials and Methods

The University of Agriculture Faisalabad's Plant Breeding and Genetics Department was the site of the current investigation. The study included eight distinct wheat genotypes with varying degrees of drought tolerance (Table 1). Since no endangered species were involved in the investigation, there was no requirement for a permit. Using PEG-6000, a study using two distinct osmotic potentials, namely 0 and 0.6 MPa, was conducted [37]. In contrast to the control treatment, which employed distilled water, the necessary amount of PEG-6000 was combined with the distilled water to create the solutions of -0.6 MPa. [37]. In Petri dishes (150×15 mm), three replicates of 25 seeds were sterilized (with 5% sodium hypochlorite) and allowed to sprout between two layers of Whatman No. 1 filter paper. Pouring 10 ml of the treatment solution or distilled water over the filter paper was followed by dosing the solution or water as needed.

To avoid evaporation, the Parafilm was used to seal the Petri dishes. For 10 days, seeds were incubated at 20 ± 2 ˚C with a 12-hour light/dark cycle. For ten days, the seed germination percentage was calculated after being recorded every 24 hours. Once the radicle had grown to a length of 1-2 mm, the seed was considered to have "germinated.

In plastic pots (97 x 165 x 90 mm) filled with a 1:3 mixture of sand and peat, a seedling growth experiment was conducted. Experiment was conducted in randomized complete block design. Ten seeds were sown in each of the three replications of the pots and placed in screen house of department. In order to produce osmatic potentials of 0, and -0.6 MPa, seeds were planted 3 cm deep, and pots were watered with PEG-6000 solutions.

The temperature was recorded 25°C and 70–80% relative humidity. When the sprouting radicle reached the soil's surface, the seed or seedling was regarded to have emerged. After twenty days various growth characteristics including root length, shoot length, fresh shoot weight dried shoot weight, root-shoot ratio and relative water contents were measured. Relative water contents were calculated using following formula:

RWC% = [(Fresh shoot weight–Dry shoot weight) / (Turgid weight–Dry shoot weight)] × 100.

The chlorophyll index was measured with SPAD meter and expressed as SPAD values. All computations were made on XLSTAT add-in of Microsoft Excel program.

Results and Discussion

PEG-induced drought stress significantly altered seed germination percentage, root and shoot length, fresh and dry weights of shoot, root:shoot ratio and chlorophyll contents. Overall, the highest seed germination percentage (92.2%) was recorded for genotype 'Bhakkar star' at 0 MPa and 70.1% at -0.6 MPa, whereas genotype 'Dilkash-20' resulted in the lowest (80.1%) at 0 MPa and 48.2 at -0.6 MPa seed germination (Table 3 and Table 4). Similarly, the highest root length at 0 MPa (28.3 cm) and the lowest (13.1 cm) root length was noted for the genotypes 'Bhakkat star' and 'Dilkash-20', respectively (Table 3). In MPa -0.6 the highest root length was Pakistan -13 (44.8 cm) and lowest indicated by Dilkash-20 (32.0 cm). The highest shoot length at 0 MPa was found in Bhakkar star (40.3 cm) and lowest was indicated by Dilkash-20 (20.0 cm). At -0.6 MPa longest shoot was observed in Bhakkar star (28.0 cm) and lowest was measured in Dilkash-20 (13.3 cm). Highest fresh shoot weight was measured in Bhakkat star at 0 MPa (0.51 g) and lowest was found in Dilkash (0.35 g). At -0.6 MPa Bhakkar star manifested highest shoot weight (0.43 g) and lowest was measured in Dilkash-20 (0.23 g). Dry shoot weight was recorded of eight commercials varieties and observed that highest value at 0 MPa was indicated by Bhakkar star (0.24 g) and lowest value manifested by Dilkash-20 (0.9 g). At -0.6 MPa highest value of dry shoot weight was indicated Bhakkat star (0.28 g) and lowest value measured in Dilkash-20 (0.11 g). At 0 MPa highest root shoot ratio was observed in Akbar -10 (0.86) and lowest was observed in Dilkash-20 (0.66). At -0.6 MPa results manifested that highest root shoot ratio was found in Fakhar e Bhakkar (2.5) and lowest was observed in Bhakkar star (1.3). Highest value of relative water contents was observed Bhakkar star (73.4%) and lowest found in Dilkash-20 (56.4%) at 0 MPa. At -0.6 MPa highest value of relative water contents were measured in Bhakkar star (60.3) and lowest observed in Dilkash-20 (29.9). Chlorophyll contents were measured with SPAD and found that highest value at 0 MPa was observed in Bhakkar star (41.8) and lowest was found in Dilkash-20 (21.2). At -0.6 MPa highest value of SPAD was measured in Bhakkar star (34.8) and lowest recorded in Dilkash-20 (17.2). Mean values of physio-morphic traits presented in (Table3 and Table 4).

Regarding genotypes by drought stress interaction, all genotypes resulted in 100% seed germination under control treatment; however, seed germination recorded a significant decrease. The genotype 'Bhakkar star' with control treatment recorded the highest values for seed germination percentage, root and shoot length, fresh and dry shoot weights relative water contents and chlorophyll contetns, whereas the lowest values for these traits were noted for the genotypes 'Dilkash-20'. At -0.6 MPa osmotic potential (Table 4) genotypes 'Bhakkar star' and 'Markaz-19' better tolerated increasing level of drought stress compared to the rest of the genotypes included in the study, whereas genotypes 'Dilkash-20' and 'Fakhr e Bhakkar' proved as the most sensitive genotypes. The decrease in seed germination percentage, root and shoot length, fresh and dry weights of shoot, relative water contents, chlorophyll contents, and increase in root:shoot ratio was significantly altered by individual and interactive effects of genotypes and PEG-induced drought stress levels.

In order to indentify the wheat genotypes desirable for breeding program for development of water deficit tolerance and high yielding cultivars, root length is distinguish and appropriate trait for choice (Ginkel and Ortiz, 2018)). Wheat plant has two types of root systems. Seminal root system starts right after germination. After germination adventious roots that, appear from the basal nodes. When seed germinates root bursts through coleorhizae and followed be emergence of 4-5 lateral seminal roots. Nodal roots appear with tillering (Ginkel and Ortiz, 2018). In osmotic stress plant adjusts itself through changing its different pathways (Wahid *et al.*, 2007; Chaves *et al.*, 2009). It illustrated that environmental changing can affect the genotypic behavior of plants for all the considered morphological and physiological attributes. Plants cannot maintain moisture contents of leaf without constant availability of water and during water unavailability, leaf moisture contents dwindle. Consequently thrash of turgor pressure (Lonbani and Arzani, 2011). During water shortage wheat cultivars produce deeper roots (Siddig *et al.*, 2013) and those plant which have capability to grow longer and deeper roots can survive well. Therefore the performance of genotypes which indicated longer root length also performed fine in water deficit conditions. Shoot acts as best source of sink for plant therefore it is very critical attribute for plant during water deficit stress. Shoot length is most drought affected parameter and decrease significantly with increasing water shortage (Ahmad *et al.*, 2013). In order to maintain the root shoot ratio during water deficit conditions, it is also very important for the plant to maintain the shoot length (Taiz and Zeiger., 2014) because during water shortage, plant use all of its reserve food for the root growth to elongate the roots because with deeper roots plant can extract water from the depth of the soil. In this process shoot indicates stunted growth. Those genotypes which indicated longer shoot length under water shortage environment should be considered as water deficit tolerant because they are least effected by water shortage conditions. In wheat genotypes which show maximum resistant to change in root shoot ratio proved to be maximum tolerant against drought (Rauf *et al.*, 2006). In morphological trait, root shoot ratio Bhakkat star indicated maximum resistant against change in root shoot ratio. Limited water supply reduces biomass of plants especially at seedling stage (Mujtaba *et al.*, 2016). Khakwani *et al.*, (2011) also documented decline in shoot fresh weight of wheat seedling due to water shortage. Water shortage is decline of available water, in response plant concentrated available solutes, e.g. carbohydrates and proline to take water and maintain water potential through osmotic regulation (Martin *et al.*, 1993). Osmotic regulation helps plant in growth and development in water dearth milieu (Pessarkli, 1999). Decrease of RWC% resulted in closing of stomata which ultimately decrease in rate of photosynthesis (Cornic, 2000). High percentage of relative water contents increase chances of survival for plant in water deficit environment (Schonfeld, *et al.*, 1988). Moderate to rigorous water deficit conditions effect plant's morphological and physiological traits. Bilal *et al.* (2015) confirmed relative water contents as excellent criteria for selection against drought stress. The ability of plant to survive in stern water deficit conditions depends on its relative water contents (Larabi and Mekliche, 2004). Relative water contents proposed as most important indicator of plant water status as compared to any other (Almeselmani *et al.*, 2011). Wheat cultivars retained maximum relative water contents are most tolerant against drought stress (Arjenaki *et al.*, 2012). In this sense RWC% are the reliable and widely used source to check the sensitivity and tolerance of plant against drought (Liu *et al.*, 2013). Final investigation indicated that Bhakkar star and Markaz-19 were in leading wheat genotypes for relative water contents.

Conclusion

The commercial wheat cultivar Bhakkar star among eight commercial wheat varieties maintained root-shoot ratio and RWC% and performed consistently in osmotic stress conditions. These genotypes should include in developing new elite drought tolerant commercial varieties. These commercial varieties can be directly recommended for arid and semi-arid regions.

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Sr.NO	**Genotype**
1	Pakistan 2013
2	Markaz-19
3	Fakhr e Bhakkar
4	Bhkkar Star
5	Dilkash-20
6	Subhani-21
7	MH-21
8	Akbar 2019

Table 2: Mean Square values from analysis of variance of physio-morphic traits at 0 MPa and -0.6 MPa

S.O.V	D.F	RL	SL	R/S	FW	DW	RWC%
Replications	2	2.20	1.02	0.032	0.04	1.8	5.6
Genotype	7	26.93**	15.77**	0.048**	0.00051**	1.195 **	1108.6**
Treatments	1	543.53**	6436.58**	1.90**	0.383**	0.003**	19767.4**
G*T	7	3.79*	7.40**	0.00983**	0.000432**	1.34**	869.8 **
Error	30	1.38**	0.02	0.00153	0.0001**	5.358**	329.7**

Table 3: Mean values of physio-morphic traits at 0 MPa

Genotypes	GP%	RL (cm)	SL (cm)	FW (g)	DW(g)	RS Ratio	RWC%	CC
Bhkkar Star	92.2	28.3	40.3	0.51	0.24	0.70	73.4	41.8
Markaz-19	90.2	25.2	35.9	0.48	0.22	0.70	71.8	40.9
Subhani-21	90.2	25.1	31.8	0.45	0.20	0.79	69.7	35.8
MH-21	89.3	23.1	28.31	0.43	0.18	0.82	66.7	33.8
Akbar 2019	88.5	20.1	23.32	0.41	0.15	0.86	64	30.23
Pakistan 2013	83.2	18.2	21.8	0.40	0.14	0.83	63	28.5
Fakhr e Bhakkar	82.1	15.2	21.56	0.40	0.11	0.71	59.19	25.9
Dilkash-20	80.1	13.1	20.0	0.35	0.9	0.66	56.48	21.2

Table 4: Mean values of physio-morphic traits at -0.6 MPa

Genotypes	GP%	RL (cm)	SL (cm)	FW (g)	DW(g)	RS Ratio	RWC%	CC
Bhkkar Star	70.1	37.3	28.3	0.43	0.28	1.3	60.3	34.8
Markaz-19	65.2	39.9	24.2	0.40	0.24	1.6	59.6	28.5
Subhani-21	62.1	41.8	22.1	0.38	0.21	1.9	55.5	25.5
MH-21	59.2	42.2	20.1	0.35	0.19	2.1	52.4	23.1
Akbar 2019	55.1	44.1	19.1	0.31	0.17	2.3	50.1	21.2
Pakistan 2013	52.5	44.8	18.5	0.28	0.15	2.4	48.5	19.7
Fakhr e Bhakkar	50.1	38.56	15.4	0.25	0.13	2.5	45.2	18.1
Dilkash-20	48.2	32.0	13.3	0.23	0.11	2.4	29.9	17.2