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Research Article

Morphological study of cotton (*Gossypium hirsutum* L.) varieties against drought tolerance under greenhouse conditions

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Abstract
Cotton is a vital fiber and oilseed crop badly affected by soil moisture deficit stress. Screening of cotton germplasm is a prerequisite to classify the cotton varieties as a drought sensitive and tolerant. With the aim to separate the distinct genotypes, ten cotton genotypes (FH-142, FH-118, MNH-886, BH-178, VH-259, SLH-317, FH-113, IUB-13, BS-52and NIAB-878) were grown under normal moisture and moisture stress environments. One group of seedling was watered at regular intervals (control) and other group was subjected to three consecutive drought cycles. The assessment was done through different morphological parameters (root length, shoot length, root fresh weight, shoot fresh weight, root dry weight, shoot dry weight and lateral root numbers).The leaf chlorophyll contents were also evaluated. The experimental design was Complete Randomized Design (CRD) in factorial way with three replicates. The data obtained was investigated statistically at 5% probability and Least Significant Difference (LSD) test was used to isolate the significant means (treatment). The results indicated that water stress adversely reduced the values of the above stated parameters excluding root length of some varieties. It is concluded that some genotypes of cotton such as FH-113, IUB-13, BS-52 and NIAB-878 indicated best response against water-deficit environment.

Keywords: Cotton; Morphological; Screening; Seedling; Varieties

Introduction

Cotton (*Gossypium hirsutum* L.) is a highly valuable fiber crop which fulfills the fiber

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needs of half of the world [1, 2]. It is the most significant source of fiber in the presence of synthetic fibers [3, 4]. Not only its fiber but also its seed is of highly valuable economically which serves as important raw material for cooking oil, and after oil extraction the residual oil cake is an important source of protein rich feed for livestock [5]. Cotton seed consists of relatively high quality oil and protein in the ratio of 21% and 23%, respectively [6]. Plastics, rubber, pharmaceuticals, cosmetics, emulsifiers, margarine and soap are the main products of cotton seed oil [7].

Cotton is an important cash crop in the world [8]. Cotton growing farmers depend solely on the income from this cash crop for buying the things and needs of their family [9]. In spite of the fact that the cotton is the most important cash crop for a large proportion of the farmers, the farmers are unable to get more yield than 25-30% of their yield potential due to drought [8]. Out of all the stress factors which affect the crop yields in the world, drought holds a prominent place which covers 26% part [10]. When plants from a same cotton variety are passed through drought, they give 50% low yield as compared to well irrigated cotton plants of the same variety [11]. Agriculture in Pakistan suffers extensive dry spells and drought due to its high dependence on rainfall for irrigation. In recent years, rainfall pattern has changed in Pakistan resulting variation in the onset of rain and its uneven distribution in different areas of the country [12]. Adequate irrigation can increase the cotton production but most of the farmers are deprived of proper irrigation facilities in the country. For small scale farmers, growing drought resistant cotton varieties is the sole method to tackle drought. Selecting different varieties for adaptation in different water deficit areas is the main strategy to develop drought tolerant varieties in breeding programs [13].

Plant Breeders have worked hardly to synthesize drought tolerant cotton varieties i.e., the characteristics of roots play major role in the survival of a plant in drought conditions and root growth is very less affected by the drought conditions but plant height, yield and shoot growth rate are retarded in water deficiency. Plants avoid water deficiency by elongating their root deeper in the soil to reach underground water table [14]. The root elongation is much more at moderate drought but severe drought root length start decreasing. When cotton seedlings are subjected to water deficiency they show elongation in roots but reduction in its diameter [14]. The difference in the measurements of root and shoot length of *G. hirsutum* seedlings shows the difference in the response of different varieties/lines to the water stress condition [15]. The growth of the root in fact indicates the response of a plant to water deficiency [14]. Considerable variation has been observed in tap root length and number of lateral root among exotic germplasm. Useful genetic variation in root growth parameters which are root length (RL), lateral root number (LRN), root fresh weight (RFW), lateral root dry weight (LRDW) and total root dry weight (TRDW) has been observed in the day-neutral converted race stocks (CRS) accessions [16]. The Considerable genetic variability for dry matter accumulation, heat tolerance and root growth exists in the foreign germplasm of *G.hirsutum*; and that vigorous root growth and number of lateral roots play important role in the adaptation of a cotton plant in water stress conditions [17]. The prior knowledge of correlation among traits discussed above is necessary to start breeding program as it provides the chance to a desirable trait to be selected [18]. There is significant positive correlation among root length, lateral root number, total dry root weight, and shoot dry weight [19].

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As there is no information about the cotton genotypes grown in Pakistan regarding the performance in water stress conditions so, there is dire need to screen the cotton genotypes for drought tolerance. This research was conducted to identify drought tolerant cotton varieties out of 10 genotypes which could show vigorous growth and give maximum productivity during dry spells.

Materials and methods

The study was conducted during kharif season in 2017 at Green House of University of Agriculture Faisalabad (altitude 184 m, latitude 31.40º N, longitude 73.05º E). Ten varieties (FH-142, FH-118, MNH-886, BH-178, VH-259, SLH-317, FH-113, IUB-13, BS-52 and NIAB-878) were planted in plastic tubes 130 x 36 cm size. These plastic tubes were subjected in the PVC pipes 120 x 30 cm size to keep straighten the plastic tubes. Plastic tubes were filled with fine sand (easy to wash roots) to grow the cotton seedlings. Ten tubes of each genotype were planted by seeding five seeds per tube and thinning of seedlings to one plant per tube were carried out 15 days post emergence. The temperature kept was 30-35˚C and 28-32˚C with 50-55% and 60-70% relative humidity at day and night, respectively. The plants were grown under water stressed and non-stressed conditions. Each tube was watered in equal amount and fertilized by NPK (20-20-20+TE) Faster (Italy), Agrium Enterprises, Multan for normal growth. Fertilization was performed by aspiration during watering. All plants in tubes were watered and fertilized in accordance with the procedure described above until the plants reached the first true leaf stage. Subsequently, the tubes were distributed randomly in two groups, each containing five tubes of each genotype. One group of seedling was watered at regular intervals (control) and other group was subjected to three consecutive drought cycles. This drought stressed regime was initiated by first with holding water when the plants reached the first true leaf stage. Each drought cycle contained withholding water for 15 days. Plants were watered to field capacity 12h after visual signs of wilting. At the end of third drought cycle from each group plants were harvested. Roots were washed free of sand and then spread on paper sheet for determination of the following parameters:

1). Root length (cm) and shoot length was measured with the help of meter rod. 2). Root fresh weight and shoot fresh weight (g plant $^{-1}$): Roots were cut at the base of the shoot, and then root fresh weight (g) was taken. Furthermore, shoots were separated from roots to calculate shoot fresh weight (g). 3). Root dry weight (g plant $^{-1}$): Root material was placed in an oven at 70ºC for 72 hours to get constant dry weight then root dry weight (g) was calculated. 4). Shoot dry weight (g plant $^{-1}$): Shoot material was placed in an oven at 70ºC for 72 hours to get constant dry weight then root dry weight (g) was calculated. 6): Lateral root number (LRN)

7): Leaf chlorophyll contents (SPAD value): Chlorophyll contents of the leaves were determined by using SPAD instrument (model SPAD-502; Minolta Corp., Ramsey, NJ)

Statistical analysis: The recorded data was statistically analyzed via Fisher's analysis of variance (ANOVA) technique. LSD test was used ($p \leq 0.05$) to compare significant treatments means using Statistic version 8.1(Analytical Software ©, 1985-2005).

Results

The observed data for root and shoot length of ten cotton varieties indicated that drought caused considerably decline in roots and shoots length of cotton varieties. However, some varieties showed good response against water stress. Under drought conditions, BS-52 (128.53 cm) showed tremendous increase in root length followed by (124 cm), IUB-13 (111.83), FH-113 (118.20 cm), SLH-317 (112.37cm), BH-178 (106.77cm), MNH-886 (97.67cm) FH-118 (93.33cm), VH-259 (91.67) and FH-142 (85.80cm) showing

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drought tolerance characteristics. Longest root length was observed for BS-52 (132.47 cm), NIAB-878 (126.03), FH-113 (124.17cm), IUB-13 (121.73 cm) while grown under irrigated condition. Shoot length shows the growth of plants. Higher the shoot length, means more photosynthates accumulation due to extensive canopy. Highest shoot length was observed in genotype BS-52 (23.667 cm) under water deficit environment. However, root and shoot length of different genotypes are more under well-watered environment than drought conditions because nutrients and water uptake reduced when soil moisture is less. Roots fresh weight of ten cotton genotypes grown under drought condition were less as compared to the genotypes grown under normal condition. However, the fresh weight of root and shoot were significantly higher in genotype BS-52 (23.507 cm) under well-watered and water deficit environment. Less root and shoot fresh weights were observed in FH-142 and VH-259 (Table 1).

Table1. Effect of water deficit stress (DD) on root length, shoot length, root fresh weight and shoot fresh weight of ten cotton varieties

Cotton Varieties	Root length (cm)		Shoot length (cm)		Root fresh weight (g)		Shoot fresh weight (g)	
	WW	**DD**	**WW**	**DD**	**WW**	**DD**	**WW**	**DD**
FH-142	85.40 M	85.80 LM	17.500 KLM	17.367 KLM	12.313 GHI	12.167 HI	14.0333 HI	12.7800 K
FH-118	101.63 HI	93.33 JK	21.400 H	16.533 LM	13.330 FGH	13.043 FGHI	14.4333 GH	13.3533 J
MNH-886	106.07 GH	97.67 IJ	19.333 IJ	18.167 JKL	13.800 FG	13.403 FGH	15.2633 E	13.9733 HI
BH-178	91.80 JK	106.77 FGH	21.600 GH	16.367 M	12.617 GHI	12.563 GHI	14.2800 GHI	13.0400 J
VH-259	84.00 M	91.67 KL	19.000 JK	18.033 JKLM	12.077 HI	11.810 I	14.0667 HI	12.7833 K
SLH-317	106.90 FGH	112.37 EF	25.033 DE	20.967 HI	14.300 F	14.240 F	16.4633 D	13.9033 I
FH-113	124.17 BCD	118.20 DE	29.667 B	23.167 FG	32.727 AB	19.267 D	17.8267 B	17.2333 C
IUB-13	111.83 FG	121.73 CD	26.933 C	21.033 HI	18.050 D	16.110 E	16.8367 CD	14.6433 FG
BS-52	132.47 A	128.53 AB	31.500 A	23.667 EF	33.507 A	31.433 BC	18.8200 A	19.0467 A
NIAB-878	126.03 BC	124 BCD	25.767 CD	21.533 GH	31.000 C	18.123 D	17.9267 B	15.0200 EF

Dry root weight, dry shoot weight, lateral root numbers and chlorophyll contents of ten cotton cultivars represent the health and growth status of plant. More dry weight were detected in BS-52 (10.047 g) and FH-113 (7.560 g) grown under normal environment as compared to these genotypes which were grown under drought environment. Overall, BS-52, FH-113, NIAB-878 and IUB-13 performed best both under drought as well as irrigated conditions. More the number of lateral roots, more will be water and nutrients uptake and plants ability to withstand the drought environment. A decrease in chlorophyll contents of leaf reduces the absorption of solar radiation that resulted into decrease in photosynthetic efficiency which effects overall growth of plants. Higher

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chlorophyll contents were found in BS-52 (52.133), FH-113 (46.967), IUB-13 (41.867) and NIAB-878 (45.100) under drought stress environment while FH-118 (36.433) and FH-142 (32.533) showed less chlorophyll contents (Table 2).

Table 2. Effect of water deficit stress (DD) on root dry weight, shoot dry weight, lateral root numbers and chlorophyll contents of eight cotton varieties

Cotton Varieties	Root dry weight (g)		Shoot dry weight (g)		Lateral root numbers		Chlorophyll contents (SPAD Value)	
	WW	**DD**	**WW**	**DD**	**WW**	**DD**	**WW**	**DD**
FH-142	1.950 GHI	0.407 M	2.1867 KL	1.5000 N	9.767 M	6.500 Q	39.667 FGH	32.533 I
FH-118	1.570 IJKL	1.187 IJKLM	3.7067 GH	2.0533 L	14.800 HI	10.200 M	40.733 EFG	36.433 GHI
MNH-886	1.843 HIJK	1.567 IJKL	4.0733 F	2.3433 JK	14.033 GH	10.633 L	45.433 DE	39.633 FGH
BH-178	1.567 IJKL	0.937 JKLM	4.8000 G	1.7300 M	14.633 I	9.300 O	42.467 DEF	36.433 GHI
VH-259	0.890 KLM	0.690 LM	3.5333 H	1.7033 MN	11.133 K	7.367 P	37.900 FGH	34.533 HI
SLH-317	2.693 EFGH	1.860 HIJ	4.2833 EF	2.5500 J	14.233 G	11.667 J	45.233 DE	42.600 DEF
FH-113	7.560 B	3.287 DE	5.0367 B	4.6600 C	15.400 E	21.000 B	56.067 B	46.967 CD
IUB-13	2.140 FGHI	3.000 EF	4.4333 DE	3.0167 I	14.727 F	11.767 J	55.000 B	41.867 DEF
BS-52	10.047 A	4.193 D	6.000 A	5.0033 B	22.067 A	15.667 D	65.000 A	52.133 BC
NIAB-878	6.357 C	2.887 EFG	4.6167 CD	4.4933 CDE	17.867 C	14.900 F	53.667 B	45.100 DE

Discussion

Various reports have declared physiological and growth response of different cotton genotypes in water-deficit environment under field conditions in both humid and arid environments and in growth chamber [18, 20-22]. However, this study on morphological responses of different cotton genotypes were conducted under greenhouse conditions to assess the drought resistance genotypes. To survive the drought stress, the plants uses two key strategies of drought tolerance and drought escape. During drought escape, plant shortens its growth period by inducing early flowering. But, this reduction in growth length results in less economic yield in indeterminate plants e.g. cotton [23].

Drought tolerance is plants ability to conserve high water contents for proper functioning i.e. drought avoidance and capability of plant endure metabolic function at less water condition i.e. drought resistance. Later rarely occur in crops. The water balance in plants can maintain due to their own physiological and morphological characteristics by reducing water loss through transpiration or enhance water absorbance due to deep and extensive root system [22].

This study revealed that root length, shoot length, fresh weight and dry weight of root and shoot, lateral roots and chlorophyll

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contents of FH-142, FH-118, MNH-886, BH-178, VH-259, SLH-317, FH-113, IUB-13, BS-52 and NIAB-878 were less under drought condition as compared to these genotypes grown under irrigated condition [22, 24, 25]. The reason behind this is that the stomatal closure due to decrease in available water reduces the transpiration and carbon dioxide absorption. This leads to reduction in photosynthesis that results in low growth rate. This is also because of the diversion of energy and photosynthetic assimilates to defensive molecules to combat the water stress. The number of lateral roots offered the better yield of crop as due to a greater number of lateral roots and root mass. Plants extract more water during stress to maintain the stomatal conductance that result into higher photosynthesis. This is also related to shoot fresh and shoot dry weight of plants as more lateral roots, the more water uptake for photosynthesis and increase the fresh and dry weight of shoot [17].

Comparable to the other studies, the findings of our study also showed that the depth and growth of rooting have significant implication for plant ability to endure drought [26-30]. Deep and more lateral roots enable the plants to resist the drought by keeping higher water potential and water uptake due to their denser root system. So, some genotypes of cotton such as FH-113, IUB-13, BS-52 and NIAB-878 indicated best response against water-deficit environment.

Conclusion

Under drought condition, BS-52 (128.53 cm) showed tremendous increase in root length followed by NIAB-878 (124 cm), IUB-13 (111.83 cm), FH-113 (118.20 cm), SLH-317 (112.37 cm), BH-178 (106.77 cm), MNH-886 (97.67 cm) FH-118 (93.33 cm), VH-259 (91.67 cm) and FH-142 (85.80 cm) demonstrated the drought tolerance characteristics. Longest root length was observed for BS-52 (132.47 cm), NIAB-878 (126.03 cm), FH-113 (124.17cm), IUB-13 (121.73 cm) while grown under irrigated condition, the fresh weight of root and shoot were significantly higher in genotype BS-52 (23.507 cm) under well-watered and water deficit environment. The Less root and shoot fresh weights were observed in FH-142 and VH-259. More dry weight were detected in BS-52 (10.047 g) and FH-113 (7.560 g) grown under normal environment as compared to these genotypes which were grown under drought environment. Overall, BS-52, FH-113, NIAB-878 and IUB-13 performed best both under drought as well as irrigated condition. Higher chlorophyll contents were founded in BS-52 (52.133), FH-113 (46.967), IUB-13 (41.867) and NIAB-878 (45.100) under drought stress environment while FH-118 (36.433) and FH-142 (32.533) showed less chlorophyll contents. It is concluded that some genotypes of cotton such as FH-113, IUB-13, BS-52 and NIAB-878 indicated best response against water-deficit environment

Authors' contributions

Conceived and designed the experiment: SU Rehman & G Akbar, Performed the experiment: SU Rehman & G Akbar, Analyzed the data: A Ahmad & K Bellitürk, Contributed reagents/ materials/ analysis tools: Z Aslam & K Yilmaz, Wrote the paper: M Nadeem, U Ali & MU Ibrahim

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