Physiological traits coupled with water deficit tolerance by using multivariate analysis in cotton genotypes.

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Abstract

The water shortage is a serious hazard for food security in the world. It is the alarming situation for the plant breeders to recognize and build up drought tolerant field crops germplasm. The water deficit tolerance magnitude rely on various soil and plant associated factors such as capacity of soil to accumulate moisture, rainfall distribution, rate of evapo-transpiration and root uniqueness. The present study was done at Central Cotton Research Institute, Multan to screen cotton germplasm against water deficit by irrigating the plants at three moisture levels at seedling stage i.e., 40, 70, and 100% of the pot capacity. The data on root length, fresh root weight, shoot length, fresh shoot weight, dry root weight, dry shoot weight and ratio of root and shoot were collected of 45 days plants after exposing to water deficit. Mean values of seedling parameters revealed differences for water deficit tolerance. Cluster analysis was performed by using Ward's method (1963). Five clusters (I, II, III, IV and V) comprised of various genotypes of cotton were observed at three moisture levels. Cluster I showed the genotypes having almost maximum values of root shoot parameters while cluster V with minimum values at all the moisture levels. These results showed the presence of drought tolerance genotypes in cluster I and susceptible genotypes in cluster V. In Biplot analysis, variables and genotypes are super imposed on the plot as vectors. The genetic variation in genotypes in different cluster showed the existence of water deficit tolerance which could be applied for the evolution of drought tolerant genotypes.

Keywords: Biplot, Cluster analysis, Drought, Moisture stress.

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Introduction

Cotton is a miraculous fiber that has a variety of uses, from fashion to home stuff to medical products Cotton, all around the world is known as very most valuable and abundant produced agricultural as well as industrial crop. Drought is a meteorological expression that means lack of precipitation of adequate amount in specific time to exhaust soil water resulting injury to plants [1]. From agricultural point of view, when existing precipitation is approximately depleted in root zone to which extent that wilting may occur. Drought is a multidimensional pressure disturbing plant at many levels of their organization. Many definitions have been given based on moisture content of soil, the potential evapotranspiration, precipitation profile or their combinations [2,3]. Drought is the more damaging factor on growth and productivity of plants as compared to any other biotic and abiotic stress in arid and semi-arid areas [4]. But cotton is one of the most important cash crop of Pakistan covering 7.86 million acres area [5]. Cotton and its other products are the integral component of economy of Pakistan with approximately 60% of total foreign exchange earnings and gross domestic products up to 10% [6]. However, per acre yield is less in Pakistan as other cotton growing countries. Drought has rigorous hazard for yield and quality among many factors [7]. Drought stress minimizes nutrient uptake and distribution within the body of plant, resulting reduction in growth and yield [8]. Different mechanisms have been adopted by plants to overcome water shortage as the developing varieties against drought are more cost-effective and viable to sustain cotton plant growth under water stress conditions [9]. In cotton, species having diploid chromosome number encompass more drought tolerance feature due to their deep rooted nature [10]. Normally, plants exhibit greater drought tolerance having deep-rooted system than shallow rooted. Thus, delay in first irrigation up to forty days in cotton, due to this mechanism roots may go longer look for soil water. Therefore, root associated characteristics might be helpful in screening of cotton genotypes against water stress. Findings have been given that root parameters can be used for drought tolerance adaptation in many crops including cowpea [11] and maize [12]. In addition to this, many wheat physiological parameters might be in second-hand for cotton genotypes screening under drought namely fresh and dry biomass, boll diameter, number of bolls and boll weight [13,14].

Soil moisture availability cope the requirement of plant growth and seed cotton yield in plentiful degree [15-17]. Roots are the main source of gaining of moisture from soil. Therefore, strategy must be changed for drought response in root growth, density and size and proliferation modification [18]. In a study by Burke et al. [19] revealed in his finding the more negative effect of drought on shoot growth as compared to root growth [20], also showed drought affect in young cotton plants. He found the prior drought effect on shoot growth as root growth. Another study by Ball et al. [12] found the effect of drought on root shoot length. He observed that there is less increase root length sensitivity than shoot to water deficit. Performed experiment under drought period after 36 days sowing of with succeeding revival time [21]. There is decrease in different traits under drought than control. Thus, root growth elongation was higher up in drought than control. The study showed the increase in length of tap roots under moisture stress than control, demonstrating that there is no effect of drought on length of tap roots of cotton. The main aim of present work is to study the behaviour of different growth stages of 50 accessions in upland cotton against water deficit situation at seedling stage. The knowledge from these findings might be supportive for plant researchers to sort out genetically potential accessions for developing drought stress tolerant germplasm designed for water stress locale.

Materials and Methods

The response of 30 cotton genotypes of upland cotton to three moisture levels was conducted in glasshouse with day and night temperature of 35°C and 28°C, respectively at Central Cotton Research Institute (latitude 31°25´N, longitude 73°09´E and altitude 184.4 m from sea level), Multan (Table 1). Screening in glass house was done at three moisture levels such as 100%, 70% and 40% of the field water capacity at seedling. Seeds of accessions were sown in October, 2013 in polythene bags with measurement of 18 × 10.2 cm size, filled with 800 g silt and 100 g of farmyard manure. Before planting of seeds all polythene bags were saturated to the designed field capacity. Overnight seeds were soaked prior to planting in bags. In each experimental bag three holes with one inch depth were made and three seeds were planted in every hole. Thinning was done after germination in such a way that one bag containing three seedlings were left. A total of four hundred and fifty (450) polythene bags were prepared for three repeats. Each repeat was divided into three sets, 100% (T0) was treated as control and 70% (T1) and 40% (T2) was predicting water stressed conditions at field capacity. The bags were arranged in completely randomized design.

Irrigation and fertilization was done in seedlings till the development of first true leaf grown under three water regimes and after that, daily irrigation was done for seedlings grown under 100% water level (T0) to keep the soil at field capacity. Withholding water supply, water stress condition was developed for seedlings at 70% (T1) and 40% (T2). Soil moisture meter (HH2 Theta Probe Type, Delta-T device, Cambridge, England). At initial wilting stage (observed visually), irrigation was done to stressed plants to relieve the wilting signs but irrigation was not more to reach field capacity level. From the date of emergence, experiment was sustained for forty-five days. After uprooting, plants were cut into roots and shoots for measuring root length, root fresh weight, shoot length and shoot fresh weight. The roots and shoots were dried for 72 h at 65 ± 5°C and the dry weight was recorded. Root: shoot ratios were calculated by dividing root traits on shoot traits. The mean values of all the parameters of each treatment grown under three moisture levels were compared to study the genotypic differences. While, grouping of genotypes on the basis of their mean performance was conducted by using [22] and the biplot graphs were formulated by software SPSS.

Results

Mean values of all the seedling traits i.e., root length, fresh root weight, dry root weight, root: shoot length ratio, fresh root: shoot weight, ratio, dry root: shoot weight ratio, shoot length, fresh shoot weight, dry shoot weight are shown in Tables 1-3.

Root parameters

Root length: Perusal of mean (Tables 1-3) for all the root parameters under controlled (100% moisture) and under two moisture levels 70% and 40% of the pot capacity. It revealed that root lengths of all the accessions measured under control conditions differed from each other, and ranged from 6.77 cm for (MS-64) to 13.97 cm for (DPL-45). Under 70% moisture conditions; root lengths were markedly reduced and ranged from 4.40 cm for GS-444 to 11.70 cm for DPL-45. Under 40% moisture level average range of root length was 2.80 (GS-444) to 9.20 cm for (Tree cotton). These genotypes performed better in all conditions are considered as drought tolerant genotypes while entries showed drastic reduction in root lengths under stressed condition may be rated as susceptible to water stress.

Fresh root weight: Means for fresh root weight under controlled (100% moisture) and at two moisture levels 70% and 40% of the pot capacity are given in Tables 1-3. Fewer than 100% moisture level maximum fresh root weight was shown by Tree cotton followed by F-14, GH-11-9-75 and MPS-11 with average values of 0.536, 0.521, 0.516 and 0.489 g respectively. Similarly minimum fresh root weight was exhibited by VS-212, NIAB-112 and CIM-84 with mean values of 0.279, 0.289 and 0.294 g respectively in Table 1. Under 70% moisture level, entries Cyto-62, GS-11-9-75, CIM-86 and SLH-41 exhibiting maximum root weight 0.360, 0.358, 0.350 and 0.350 g respectively while minimum fresh root weight was shown by GS 444 (0.203 g), VS-212 (0.219 g) and NIAB-112 (0.219 g) in Table 2. Fewer than 40% moisture level, Tree cotton, F-14, BP-52 and MPS-11 revealed maximum value 0.299, 0.293, 0.289 and 0.282 g respectively while COOKER-315 (0.152 g) and GS-444 (0.155 g) showed minimum fresh root in Table 3.

Dry root weight: Based upon dry root weight, data is given in Tables 1-3, 50 accessions again appeared to respond differently to controlled and stressed conditions. At 100% moisture level maximum dry root weight was exhibited by Tree cotton (0.063 g) followed by DPL-45 (0.063 g), Cyto-62 (0.062 g), BH-176 (0.060 g) and F-14 (0.059 g) while minimum by GS-444, MS-64 and COOKER-315 with 0.047, 0.048 and 0.048 g respectively. Similarly, in DPL-45 reduction was from 0.063 g at 100% to 0.049 g at 70% moisture level followed by Tree cotton, Cyto-62 (0.062 g to 0.049 g) and MPS-11 (0.048-0.059) g. Based on this performance these entries can be marked as most tolerant genotypes. While minimum dry root weight exhibited at 70% moisture level was by MS-64 (0.039) and COOKER-315 (0.039). When 40% moisture was applied Tree cotton was most tolerant genotype with 0% reduction in weight with increase in stress level and dry root weight also remained constant with value of 0.049g at both stresses levels while GS-

Table 1. *Mean values of seedling traits of cotton accessions grown under 100% moisture level of field capacity.*

Sr. No.	Genotypes	Root Traits						Shoot Traits		
		RL	FRW	DRW	RL/SL	FRW/FSW	DRW/DSW	SL	FSW	DSW
1	GS-444	7.30	0.333	0.047	0.591	0.243	0.168	12.37	1.37	0.280
2	Cyto-124	8.43	0.386	0.058	0.548	0.213	0.148	15.40	1.81	0.394
3	DNH-105	9.00	0.377	0.057	0.518	0.168	0.116	17.37	2.24	0.493
4	CRIS-533	7.87	0.373	0.057	0.432	0.203	0.117	18.23	1.84	0.484
5	MPS-27	8.70	0.389	0.057	0.575	0.224	0.150	15.13	1.74	0.382
6	CIM-506	8.53	0.347	0.057	0.586	0.182	0.126	14.57	1.91	0.455
7	TH-112/05	9.43	0.401	0.055	0.536	0.221	0.113	17.63	1.82	0.489
8	PB-896	10.50	0.409	0.053	0.575	0.248	0.137	18.27	1.65	0.391
9	Sun-02	10.13	0.425	0.059	0.519	0.199	0.130	19.57	2.13	0.454
10	CIM-573	11.47	0.452	0.051	0.666	0.257	0.120	17.23	1.76	0.425
11	BH-176	13.70	0.465	0.060	0.731	0.207	0.104	18.73	2.24	0.574
12	CIM-591	9.40	0.315	0.056	0.556	0.219	0.151	16.93	1.44	0.375
13	NIA-80	10.30	0.359	0.050	0.634	0.216	0.136	16.23	1.66	0.370
14	CRIS-510	9.50	0.345	0.059	0.576	0.199	0.135	16.50	1.73	0.437
15	VH-300	8.90	0.318	0.056	0.476	0.156	0.124	18.70	2.04	0.453
16	VS-212	8.63	0.279	0.054	0.439	0.132	0.115	19.67	2.11	0.472
17	MPS-11	12.87	0.489	0.059	0.682	0.279	0.105	18.90	1.75	0.562
18	DPL-45	13.97	0.469	0.063	0.760	0.209	0.113	18.37	2.24	0.562
19	NIAB-112	10.37	0.289	0.054	0.597	0.129	0.115	17.37	2.24	0.474
20	CIM-608	8.17	0.295	0.050	0.498	0.139	0.119	16.40	2.12	0.420
21	IUB-2011	8.70	0.367	0.057	0.557	0.199	0.131	15.67	1.85	0.438
22	PB-38	10.27	0.443	0.049	0.613	0.281	0.127	16.77	1.57	0.388
23	CIM-534	9.60	0.318	0.052	0.586	0.156	0.117	16.40	2.04	0.447
24	CIM-612	10.13	0.439	0.051	0.649	0.249	0.153	15.60	1.77	0.334
25	CIM-473	10.40	0.418	0.051	0.672	0.272	0.158	15.50	1.54	0.322
26	L-229-29-71	9.50	0.416	0.055	0.625	0.310	0.159	15.20	1.34	0.347
27	B-452	7.80	0.468	0.058	0.487	0.256	0.127	16.07	1.83	0.459
28	Stone ville-603	8.57	0.404	0.056	0.456	0.209	0.127	18.80	1.93	0.444
29	Tree Cotton	13.40	0.536	0.063	0.727	0.240	0.113	18.43	2.24	0.553
30	BP-52	9.67	0.465	0.055	0.684	0.280	0.144	14.13	1.66	0.387
31	COOKER-312	9.87	0.337	0.058	0.599	0.200	0.147	16.47	1.68	0.395
32	RA-31-21	10.13	0.434	0.059	0.703	0.247	0.173	14.43	1.76	0.346
33	MS-64	6.77	0.364	0.048	0.561	0.250	0.177	12.07	1.46	0.273
34	CIM-84	7.43	0.294	0.049	0.424	0.137	0.117	17.57	2.14	0.416
35	AC-307	12.43	0.465	0.058	0.679	0.206	0.126	18.30	2.26	0.460
36	NIAB-78	10.10	0.351	0.057	0.650	0.181	0.141	15.53	1.94	0.405
37	GH-11-9-75	9.80	0.516	0.057	0.632	0.296	0.152	15.50	1.74	0.374
38	CIM-86	9.00	0.488	0.059	0.552	0.252	0.132	16.30	1.94	0.445
39	CIM-43	11.23	0.435	0.053	0.723	0.245	0.153	15.53	1.78	0.348
40	Karishma	9.03	0.355	0.050	0.597	0.204	0.147	15.13	1.74	0.342
41	COOKER-315	8.10	0.300	0.048	0.538	0.197	0.175	15.07	1.52	0.277
42	SLH-41	10.77	0.454	0.056	0.648	0.247	0.158	16.63	1.84	0.358
43	Cyto-62	13.07	0.476	0.062	0.766	0.214	0.113	17.10	2.22	0.552
44	CRIS-134	9.77	0.322	0.050	0.662	0.199	0.126	14.77	1.62	0.405
45	CRIS-9	10.43	0.400	0.054	0.591	0.219	0.128	17.67	1.83	0.420
46	ME-115	11.37	0.468	0.053	0.601	0.209	0.115	18.90	2.24	0.460
47	CIM-57	10.40	0.447	0.055	0.720	0.277	0.152	14.47	1.62	0.367
48	F-14	10.17	0.521	0.059	0.738	0.296	0.134	13.77	1.76	0.443
49	S-71	11.20	0.439	0.058	0.627	0.252	0.134	17.90	1.75	0.431
50	CIM-496	12.40	0.431	0.058	0.694	0.226	0.128	18.03	1.91	0.452

RL-Root Length, FRW- Fresh Root Weight; DRW- Dry Root Weight; RL/SL- root: Shoot Length Ratio, FRW/FSW- Fresh Root: Shoot Weight, Ratio, DRW/DSW-Dry Root: Shoot Weight Ratio, SL-Shoot Length, FSW-Fresh Shoot Weight, DSW-Dry Shoot Weight

444 was most susceptible (0.023 g). After applying 100% water and both moisture stress levels i.e., 70% and 40% respectively maximum average performance was exhibited by Tree cotton (0.054 g) followed by Cyto-62 (0.052 g), DPL-45 (0.052 g) and RA-31-21 (0.050 g). These entries reduced their weight about 14% after exposure to stress condition while susceptible entries reduced about 27% average weight after stress treatment and these entries include GS-444, MS-64, Cooker-315 and CIM-608 with average dry root weight of 0.034, 0.039, 0.040 and 0.041 g respectively.

Root shoot length ratio: Based upon root shoot length ratio data is given in Tables 1-3. Fewer than 100% moisture level maximum root shoot length ratio was exhibited by DPL-45 followed by F-14, BH-176 and Tree cotton with ratio of 0.760, 0.738, 0.731 and 0.727 respectively while minimum ratio was

Table 2. *Mean values of seedling traits of cotton accessions grown under 70% moisture level of field capacity.*

Sr.	Genotypes	Root Traits						Shoot Traits		
No.		RL	FRW	DRW	RL/SL	FRW/FSW	DRW/DSW	SL	FSW	DSW
1	GS-444	4.40	0.203	0.031	0.589	0.238	0.211	7.47	0.85	0.151
2	Cyto-124	6.20	0.275	0.044	0.511	0.169	0.148	12.13	1.62	0.299
3	DNH-105	6.43	0.285	0.044	0.481	0.165	0.138	13.40	1.73	0.320
4	CRIS-533	5.47	0.262	0.042	0.381	0.183	0.134	14.37	1.43	0.314
5	MPS-27	6.53	0.287	0.043	0.567	0.228	0.191	11.53	1.26	0.225
6	CIM-506	5.80	0.275	0.042	0.537	0.190	0.140	10.80	1.45	0.298
7	TH-112/05	6.47	0.311	0.040	0.510	0.231	0.136	12.67	1.35	0.295
8	PB-896	7.40	0.301	0.042	0.600	0.266	0.195	12.37	1.13	0.214
9	Sun-02	7.20	0.319	0.047	0.492	0.230	0.154	14.63	1.39	0.306
10	CIM-573	8.17	0.295	0.041	0.646	0.206	0.141	12.67	1.43	0.291
11	BH-176	11.63	0.320	0.049	0.742	0.186	0.126	15.70	1.72	0.398
12	CIM-591	6.50	0.222	0.046	0.551	0.219	0.189	11.80	1.02	0.250
13	NIA-80	6.80	0.283	0.042	0.665	0.227	0.169	10.23	1.25	0.250
14	CRIS-510	5.97	0.260	0.045	0.487	0.187	0.148	12.27	1.39	0.306
15	VH-300	6.80	0.245	0.044	0.457	0.168	0.158	14.93	1.46	0.284
16	VS-212	6.20	0.219	0.043	0.407	0.133	0.139	15.27	1.65	0.310
17	MPS-11	10.70	0.350	0.048	0.677	0.192	0.123	15.80	1.83	0.391
18	DPL-45	11.70	0.325	0.049	0.786	0.184	0.127	14.90	1.77	0.383
19	NIAB-112	7.90	0.219	0.044	0.738	0.125	0.141	10.70	1.75	0.315
20	CIM-608	6.10	0.221	0.042	0.519	0.133	0.137	11.77	1.67	0.310
21	IUB-2011	6.70	0.236	0.047	0.691	0.192	0.146	9.70	1.23	0.320
22	PB-38	7.40	0.268	0.039	0.714	0.233	0.195	10.37	1.15	0.206
23	CIM-534	6.10	0.248	0.038	0.528	0.183	0.169	11.60	1.35	0.233
24	CIM-612	6.37	0.264	0.039	0.601	0.232	0.155	10.60	1.14	0.251
25	CIM-473	6.70	0.302	0.040	0.679	0.283	0.154	9.87	1.07	0.260
26	L-229-29-71	7.47	0.337	0.041	0.728	0.370	0.202	10.27	0.92	0.210
27	B-452	6.00	0.318	0.046	0.489	0.187	0.159	12.27	1.70	0.292
28	Stone ville-603	5.93	0.301	0.046	0.459	0.244	0.186	12.93	1.23	0.245
29	Tree Cotton	11.40	0.347	0.049	0.764	0.212	0.132	14.93	1.64	0.376
30	BP-52	6.70	0.307	0.046	0.705	0.266	0.188	9.53	1.16	0.250
31	COOKER-312	7.47	0.255	0.047	0.719	0.218	0.186	10.40	1.17	0.257
32	RA-31-21	7.43	0.302	0.047	0.849	0.249	0.191	8.77	1.22	0.250
33	MS-64	5.03	0.250	0.039	0.674	0.222	0.205	7.47	1.13	0.189
34	CIM-84	5.43	0.227	0.040	0.437	0.137	0.138	12.47	1.66	0.287
35	AC-307	9.70	0.322	0.041	0.659	0.181	0.153	14.73	1.78	0.270
36	NIAB-78	6.80	0.278	0.041	0.619	0.206	0.162	11.00	1.35	0.260
37	GH-11-9-75	7.43	0.358	0.041	0.713	0.287	0.179	10.43	1.25	0.235
38	CIM-86	6.20	0.356	0.046	0.518	0.257	0.164	11.97	1.39	0.281
39	CIM-43	8.50	0.316	0.042	0.753	0.237	0.145	11.30	1.34	0.290
40	Karishma	6.70	0.264	0.042	0.698	0.230	0.163	9.60	1.15	0.262
41	COOKER-315	5.10	0.219	0.039	0.574	0.239	0.205	8.90	0.92	0.190
42	SLH-41	6.77	0.350	0.044	0.577	0.259	0.135	11.73	1.35	0.325
43	Cyto-62	10.80	0.360	0.049	0.748	0.194	0.126	14.43	1.86	0.390
44	CRIS-134	6.17	0.283	0.041	0.642	0.233	0.141	9.60	1.24	0.292
45	CRIS-9	7.87	0.310	0.043	0.587	0.332	0.164	13.40	0.93	0.263
46	ME-115	8.70	0.275	0.043	0.596	0.168	0.195	14.60	1.64	0.223
47	CIM-57	7.20	0.232	0.045	0.683	0.172	0.182	10.53	1.35	0.253
48	F-14	7.40	0.348	0.047	0.884	0.239	0.170	8.37	1.46	0.276
49	S-71	8.20	0.250	0.043	0.712	0.204	0.189	11.53	1.23	0.232
50	CIM-496	8.50	0.329	0.046	0.642	0.245	0.168	13.27	1.34	0.274

RL-Root Length, FRW- Fresh Root Weight; DRW- Dry Root Weight; RL/SL- Root: Shoot Length Ratio, FRW/FSW- Fresh Root: Shoot Weight, Ratio, DRW/DSW-Dry Root: Shoot Weight Ratio, SL-Shoot Length, FSW-Fresh Shoot Weight, DSW-Dry Shoot Weight

shown by CRIS-533 (0.432) and CIM-84 (0.424) as in Table 1. Under controlled condition at 70% moisture level maximum ratio was shown by F-14 (0.884), RA-31-21 (0.849), DPL-45 (0.786) and Tree cotton (0.764) while minimum ratio was given by VS-212 (0.407) and CRIS (0.381) given in Table 2. At 40% irrigation level maximum ratio possessing entries were F-14 (1.106) and ME-15 (0.982). Similarly minimum ratios were obtained from Stone valley-603 and CRIS-533 with root shoot length ratio of 0.392 and 0.399 respectively in Table 3.

Fresh root shoot weight ratio: In Tables 1-3, all accessions again appeared to respond differently both under normal and stressed conditions based upon fresh root shoot weight ratio Under normal conditions fresh root shoot weight ratio range was 0.129 (NIAB-111) to 0.310 (L-229-29-71) at 70% moisture level fresh root shoot weight ratio ranged from 0.125 (NIAB-112) to -0.370 (L-229-29-71) and at 40% moisture level, fresh root shoot weight ratio ranged from 0.140 (CYTO-620 to 0.432 (CRIS-9) as shown in Tables 1-3 respectively. At maximum

***Table 3.** Mean values of seedling traits of cotton accessions grown under 40% moisture level of field capacity.*

Sr. No.	Genotypes	Root Traits						Shoot Traits		
		RL	FRW	DRW	RL/SL	FRW/FSW	DRW/DSW	SL	FSW	DSW
1	GS-444	2.80	0.155	0.023	0.675	0.273	0.222	4.17	0.57	0.103
2	Cyto-124	4.93	0.241	0.034	0.617	0.222	0.202	8.00	1.22	0.175
3	DNH-105	4.13	0.241	0.045	0.399	0.179	0.249	10.37	1.35	0.183
4	CRIS-533	4.07	0.213	0.040	0.399	0.175	0.209	10.20	1.22	0.193
5	MPS-27	5.17	0.208	0.033	0.587	0.180	0.174	8.80	1.15	0.189
6	CIM-506	4.70	0.215	0.037	0.553	0.237	0.186	8.50	0.91	0.202
7	TH-112/05	3.87	0.219	0.037	0.455	0.221	0.201	8.50	0.99	0.181
8	PB-896	4.73	0.266	0.033	0.585	0.272	0.206	8.10	0.99	0.165
9	Sun-02	3.93	0.268	0.040	0.409	0.314	0.202	9.63	0.86	0.198
10	CIM-573	5.40	0.175	0.034	0.724	0.151	0.160	7.47	1.16	0.215
11	BH-176	8.93	0.271	0.040	0.860	0.183	0.123	10.40	1.48	0.332
12	CIM-591	4.37	0.205	0.040	0.555	0.257	0.198	7.83	0.80	0.203
13	NIA-80	4.80	0.242	0.035	0.735	0.275	0.183	6.53	0.88	0.200
14	CRIS-510	4.23	0.208	0.040	0.510	0.230	0.239	8.30	0.91	0.170
15	VH-300	5.00	0.214	0.040	0.531	0.253	0.194	9.43	0.85	0.208
16	VS-212	5.10	0.169	0.039	0.524	0.171	0.168	9.73	0.99	0.240
17	MPS-11	8.60	0.282	0.044	0.820	0.220	0.134	10.50	1.28	0.328
18	DPL-45	9.00	0.244	0.044	0.882	0.192	0.133	10.20	1.27	0.338
19	NIAB-112	5.50	0.189	0.038	0.764	0.162	0.155	7.20	1.17	0.250
20	CIM-608	4.27	0.163	0.033	0.615	0.194	0.158	6.97	0.85	0.207
21	IUB-2011	4.00	0.218	0.040	0.586	0.277	0.190	6.83	0.79	0.212
22	PB-38	4.87	0.217	0.034	0.669	0.264	0.203	7.30	0.82	0.168
23	CIM-534	5.10	0.190	0.036	0.680	0.222	0.182	7.50	0.86	0.198
24	CIM-612	4.30	0.253	0.036	0.580	0.296	0.183	7.43	0.86	0.196
25	CIM-473	4.20	0.228	0.038	0.577	0.306	0.225	7.30	0.75	0.170
26	L-229-29-71	5.93	0.211	0.033	0.886	0.334	0.189	6.70	0.63	0.175
27	B-452	4.40	0.250	0.032	0.564	0.270	0.164	7.80	0.93	0.194
28	Stone ville-603	3.43	0.220	0.043	0.392	0.358	0.226	8.77	0.62	0.190
29	Tree Cotton	9.20	0.299	0.049	0.800	0.266	0.141	11.50	1.13	0.351
30	BP-52	4.80	0.289	0.044	0.663	0.343	0.214	7.23	0.84	0.204
31	COOKER-312	5.37	0.195	0.043	0.661	0.229	0.234	8.13	0.85	0.182
32	RA-31-21	4.57	0.260	0.042	0.703	0.309	0.200	6.50	0.84	0.213
33	MS-64	3.17	0.177	0.032	0.781	0.210	0.300	4.07	0.85	0.107
34	CIM-84	3.93	0.205	0.041	0.472	0.178	0.240	8.33	1.15	0.170
35	AC-307	6.50	0.271	0.038	0.753	0.221	0.193	8.63	1.23	0.195
36	NIAB-78	5.90	0.228	0.039	0.820	0.237	0.233	7.20	0.96	0.166
37	GH-11-9-75	5.80	0.267	0.036	0.777	0.280	0.186	7.50	0.96	0.193
38	CIM-86	4.50	0.219	0.041	0.538	0.258	0.194	8.37	0.85	0.211
39	CIM-43	5.80	0.183	0.034	0.746	0.156	0.179	7.80	1.18	0.193
40	Karishma	5.03	0.219	0.036	0.694	0.230	0.171	7.27	0.95	0.214
41	COOKER-315	3.07	0.152	0.034	0.615	0.230	0.322	5.00	0.66	0.109
42	SLH-41	4.87	0.266	0.041	0.569	0.357	0.236	8.57	0.75	0.175
43	Cyto-62	7.90	0.231	0.046	0.868	0.140	0.132	9.10	1.65	0.350
44	CRIS-134	4.23	0.197	0.039	0.538	0.235	0.190	7.87	0.84	0.206
45	CRIS-9	6.00	0.278	0.041	0.657	0.432	0.212	9.13	0.64	0.192
46	ME-115	7.27	0.178	0.035	0.982	0.166	0.190	7.40	1.07	0.185
47	CIM-57	4.27	0.160	0.042	0.569	0.169	0.161	7.50	0.95	0.261
48	F-14	6.33	0.293	0.042	1.106	0.355	0.238	5.73	0.83	0.181
49	S-71	5.00	0.170	0.032	0.656	0.164	0.170	7.63	1.04	0.186
50	CIM-496	5.63	0.272	0.042	0.600	0.291	0.215	9.40	0.94	0.196

RL-Root Length, FRW- Fresh Root Weight; DRW- Dry Root Weight; RL/SL- Root: Shoot Length Ratio, FRW/FSW- Fresh Root: Shoot Weight, Ratio, DRW/DSW-Dry Root: Shoot Weight Ratio, SL-Shoot Length, FSW-Fresh Shoot Weight, DSW-Dry Shoot Weight

stress level, as 40% of the field capacity, maximum fresh root shoot weight ratio was exhibited by L-229-29-71 followed by CRIS-9, F-14 and BP-52 with average values of 0.338, 0.327, 0.297 and 0.296 respectively. Similarly, minimum ratio for fresh root shoot ratio was shown by VS-212 (0.145) and NIAB-112 (0.139).

Dry root shoot weight ratio: Entries performing better under stress conditions (Tables 2 and 3) exhibited low dry root shoot weight ratio as compared to normal condition genotypes e.g., at 100% field capacity (Table 3). Under normal condition, minimum ratio was delivered by BH-176 followed by MPS-11, Cyto-62, Tree cotton and DPL-45 with average ratio of 0.104, 0.105, 0.113, 0.113 and 0.113 respectively while entries GS-444 (0.168), RA-31.21 (0.173), Cooker-315 (0.175) and MS-64 (0.177) showed maximum dry root shoot weight ratio as given in Table 1. At 70% moisture level the entries that showed

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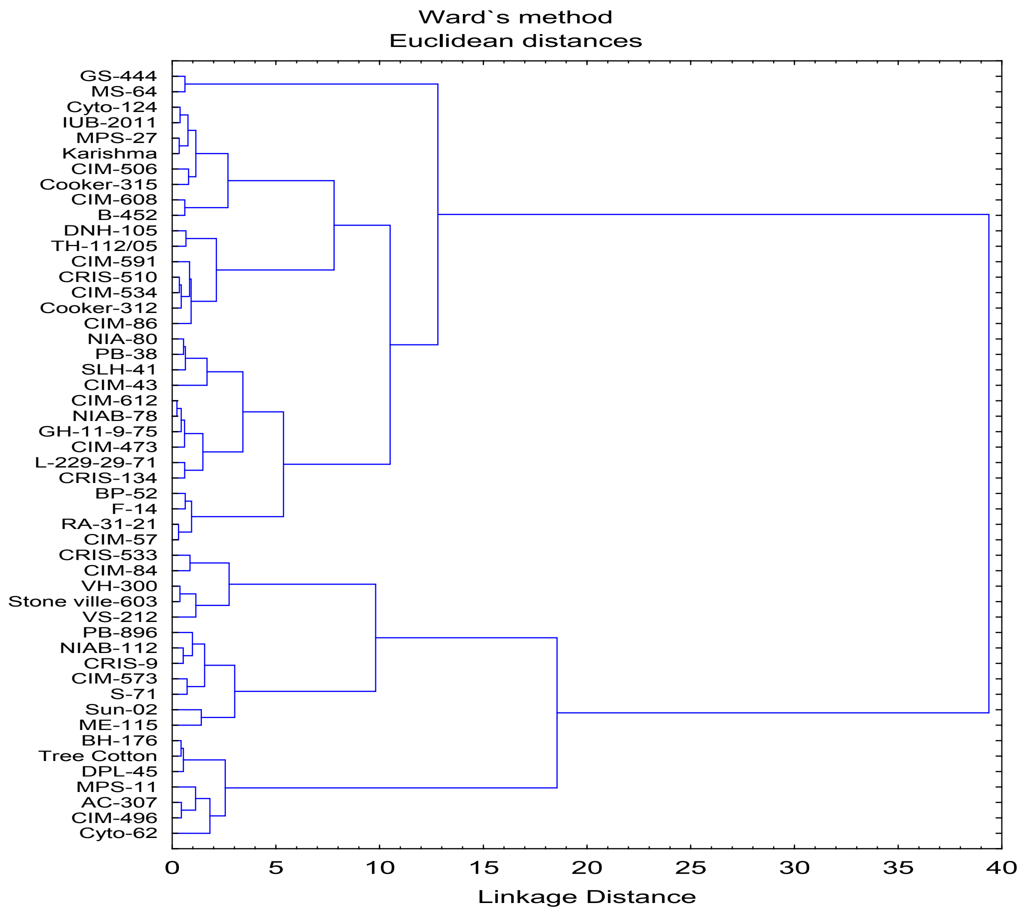

Figure 1. *Dendrogram generated by Ward's method of cluster analysis among the 50 cotton genotypes under (100% moisture level).*

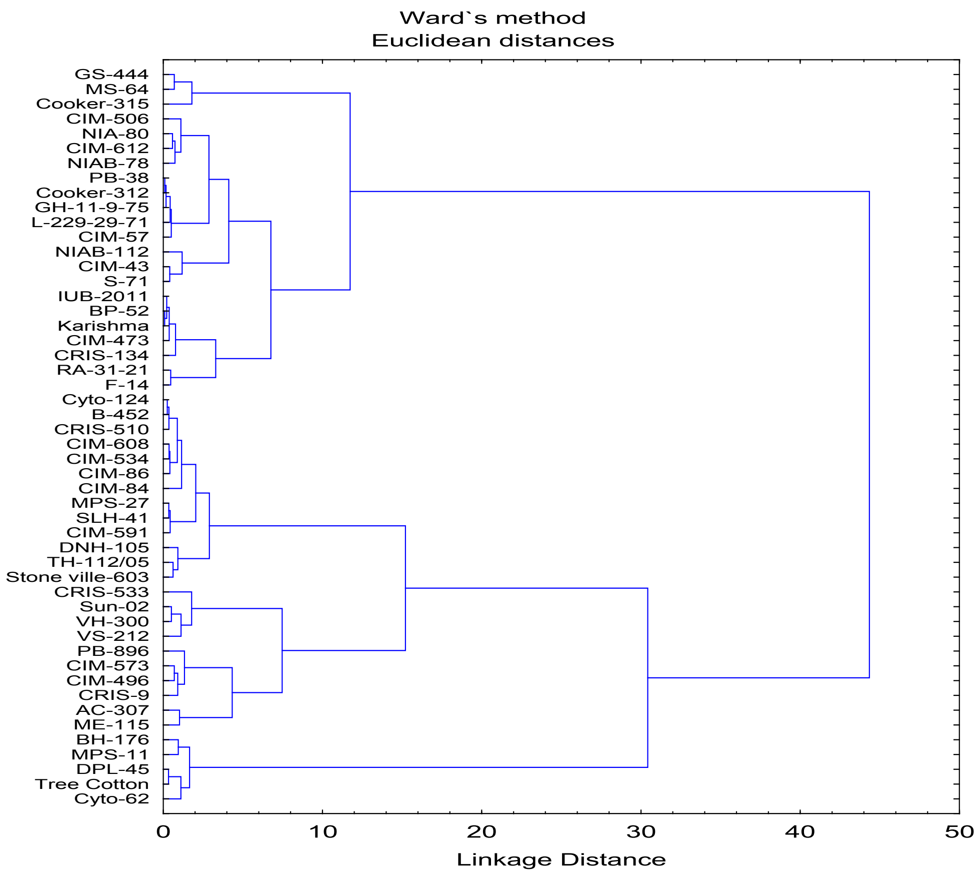

Figure 2. *Dendrogram generated by Ward's method of cluster analysis among the 50 cotton genotypes under (70% moisture level).*

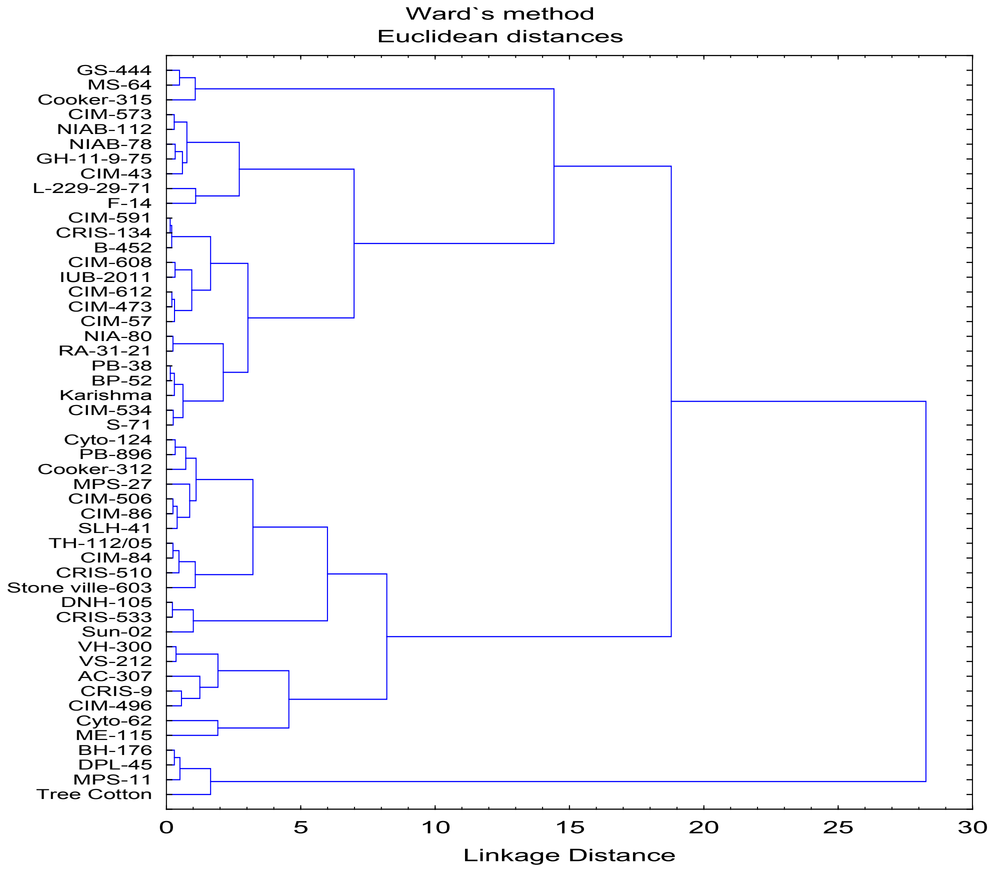

Figure 3. *Dendrogram generated by Ward's method of cluster analysis among the 50 cotton genotypes under (40% moisture level).*

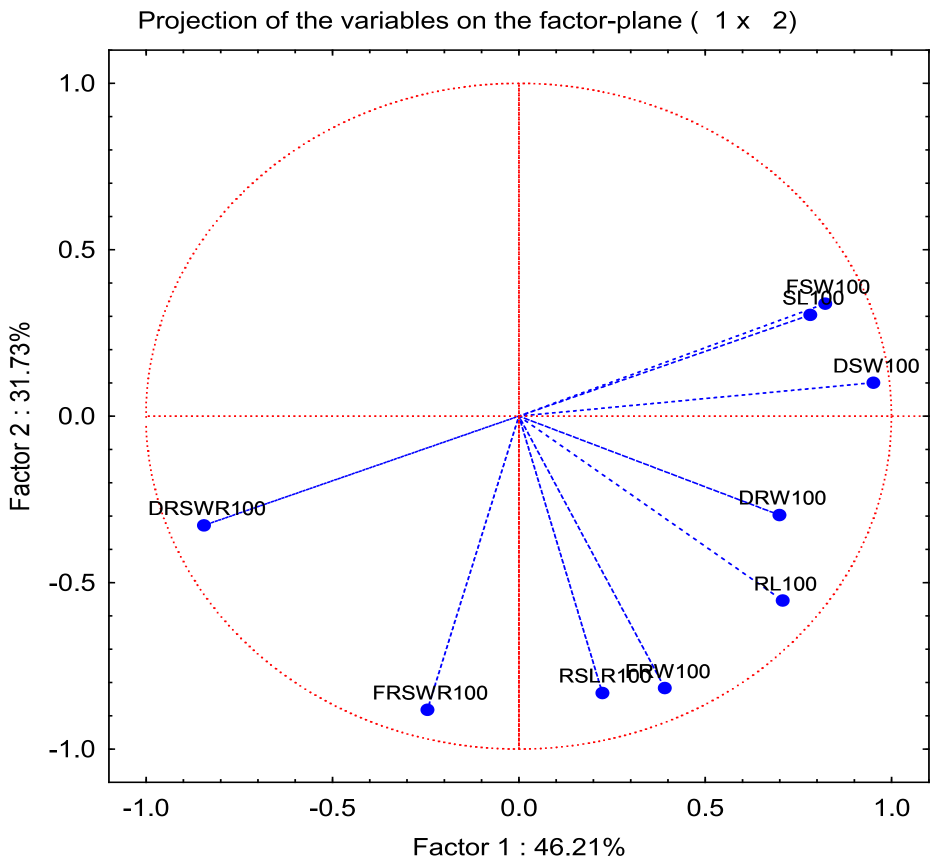

Figure 4. *Biplot between PCs 1 and 2 showing contribution of various traits of cotton accessions under (100% moisture level).*

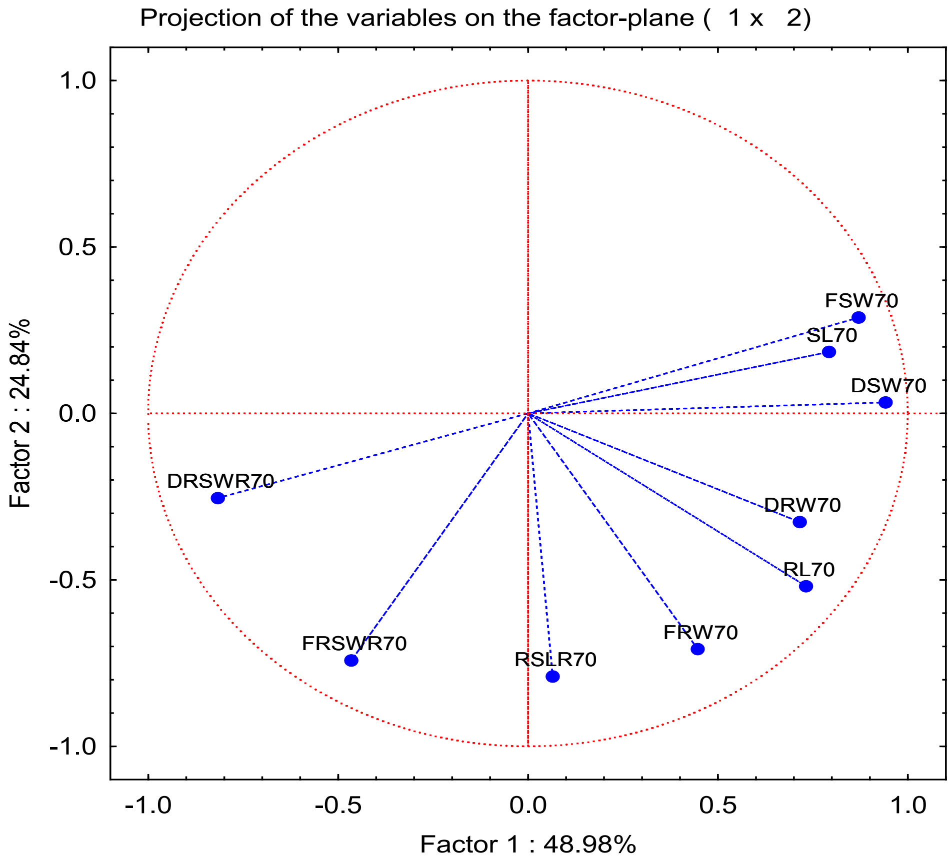

Figure 5. *Biplot between PCs 1 and 2 showing contribution of various traits of cotton accessions under (70% moisture level).*

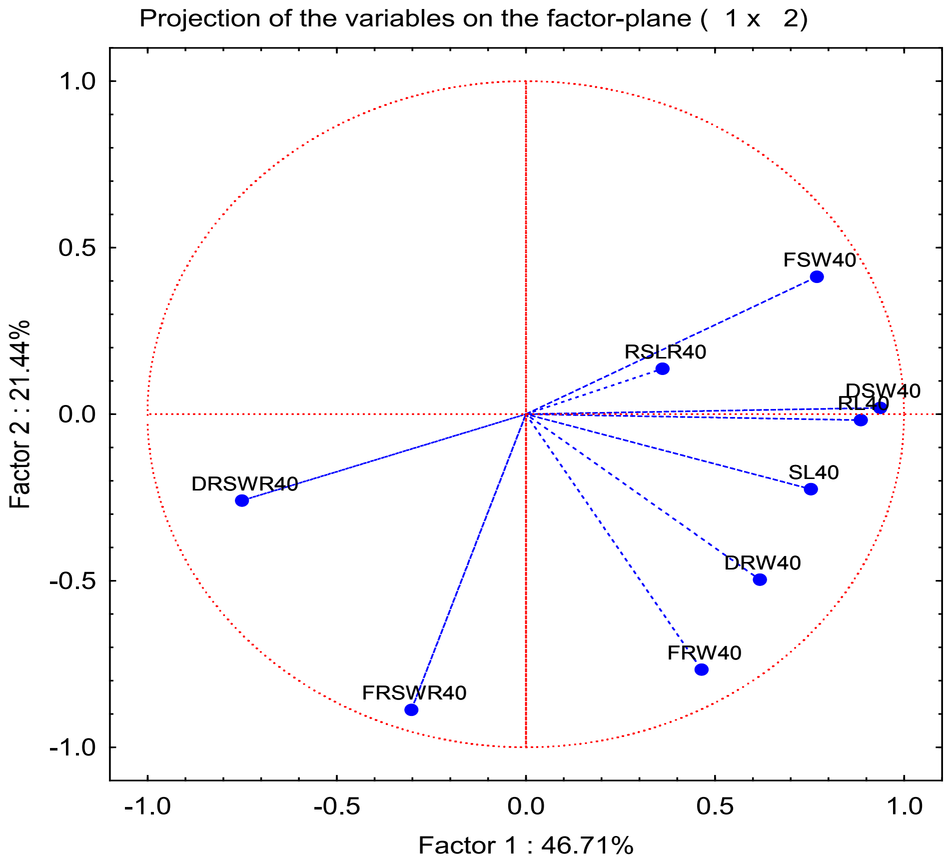

Figure 6. *Biplot between PCs 1 and 2 showing contribution of Various traits of cotton accessions under (70% moisture level).*

maximum ratios of dry root shoot weight ratio were Cooker-315, MS-64 and GS-444 with average of 0.205, 0.205 and 0.211 respectively. Minimum ratio was exhibited by MPS-11, Cyto-62, BH-176, DPL-45 and Tree cotton with ratio of 0.123, 0.126, 0.126, 0.127 and 0.132 respectively in Table 2. At 40% moisture level, entries with poor performance under maximum stress showed maximum dry root shoot weight ratio were Cooker-315 (0.322) and MS-64 (0.300) while minimum ratio was delivered by BH-176 followed by Cyto-62, DPL-45, MPS-11 and Tree cotton with value of dry root shoot weight ratio of 0.123, 0.132, 0.133, 0.134 and 0.144 respectively in Table 3.

Shoot parameters

Shoot length: Shoot length is an important parameter to study the effect of drought in cotton and many other crops. Shoot length of 50 different accessions of cotton was recorded under three different levels i.e., 100%, 70% and 40% of the pot capacity. The shoot data for all 50 genotypes showed that shoot length decreased as the moisture stress level increased.

At 100% moisture level, shoot length ranged from 12.07 cm for (MS-64) to 19.67 cm for (VS-212) in Table 1, similarly at 70% moisture levels its range was 7.47 cm (MS-64 and GS-444) to 15.80 cm (MPS-11) in Table 2 and at 40% moisture level, shoot length was 4.07 cm (MS-64) to 11.50 cm (MPS-11) in Table 3. At 100% moisture level, maximum shoot length was exhibited by VS-212, Sun-02 and MPS-11 with average value of 19.67, 19.57, 18.90 and 18.90 cm respectively. While minimum shoot length was exhibited by MPS-64 (12.07 cm) and GS-444 (12.3 cm) given in Table 1. At 70%, moisture level maximum shoot length was given by MPS-11 (15.80 cm) and BH-176 (15.70 cm) while minimum shoot length was shown by MS-64 and GS-444 with average value of 7.47 cm as in Table 2. At 40% moisture level, shoot length was 4.07 cm (MS-64) to 11.50 cm (MPS-11) in Table 3. When stress was increased by applying 2 irrigations levels genotypes which remained tolerant were Tree cotton, MPS-11, BH-176 with value of 11.50, 10.50 and 10.40 cm respectively. Similarly entries which reduced their shoot length more at this higher level of stress were MS-64, GS-44 and COOKER-315 exhibiting minimum shoot length of 4.07, 4.17 and 5.00 cm respectively.

Fresh shoot weight: Mean performance of cotton genotypes revealed that AC-307 had maximum fresh shoot weight (2.26 g) under normal conditions followed by DNH-105, BH-176 and DPL-45 (2.24 g) each while GS-444 had (1.37 g) and L-229-29-71 had minimum fresh shoot weight (1.34 g) as in Table 1. Cyto-62 and MPS-11 had maximum fresh shoot weight (1.86 g) and (1.83 g) at 70% moisture level in Table 2 while GS-444 and Cooker-315 had minimum fresh shoot weight 0.85 g and 0.92 g respectively. At 40% moisture level, it was noted that Cyto-62 had maximum fresh shoot weight followed by BH-176, DNH-105, MPS-11 and DPL-11 with average value of 1.65, 1.48, 1.35, 1.28 and 1.27 g respectively in Table 3.

Dry shoot weight: In cotton, dry shoot weight parameter is commonly used for screening cotton genotypes against drought stress. Under normal condition (Table 1) maximum dry root weight was shown by BH-176 (0.574 g), MPS-11, DPL-45 (0.562 g), Tree cotton (0.533 g) and Cyto-62 (0.552 g) while minimum dry root weight was exhibited byGS-444,COOKER-315 and MS-64 with value of 0.280, 0.277 and 0.273 g respectively. When 70% moisture level was applied, maximum dry shoot weight was given by i.e., BH-176, MPS-11, Cyto-62, DPL-45 and Tree cotton with value of 0.398, 0.391, 0.390, 0.383 and 0.376 g respectively. Minimum dry shoot weight was exhibited by COOKER-315, MS-64 and GS-444, with value of 0.190, 0.189 and 0.151 g respectively in Table 2. Similarly at 40% moisture level (Table 3) maximum dry shoot weight was predicted by Tree cotton (0.351 g), Cyto-62 (0.350 g), DPL-45 (0.338 g), BH-176 (0.332 g) and MPS-11(0.328 g) while minimum value observed by COOKER-315 (0.109 g) MS-64 (0.107) and GS-444 (0.103).

This study was done for cotton genotypes grouping with the help of multivariate analysis by classifying the germplasm into different clusters on genetic potential basis. Fifty cotton accessions were screened under stress conditions for the assessment of genetic variability at seedling stage. Cluster analysis was applied for analyzing various variables and grouped into different clusters based on similarities and dissimilarities. Dendrograms were constructed for dividing of fifty cotton accessions sown in water stressed environment. Thus, five drought tolerant accessions were screened out as lines and three drought susceptible accessions were identified as testers. Cluster analysis showed the existence of significant phonotypical diversity within fifty cotton genotypes (Figures 1-3). Phonograms were constituted based on nine seedling parameters remarkably affected by water stress. Dendrograms were divided into five clusters. It was noticed that genotypes belonging to cluster -I performed best for all of the traits including root length, fresh root weight, dry root weight; root: shoot length ratio, fresh root: shoot weight, ratio, dry root: shoot weight ratio, shoot length, fresh shoot weight, dry shoot weight. Cluster-1 was named as drought tolerant group and could be used as lines for developing drought tolerant germplasm. The cluster II, III, IV and V had 10, 13, 19 and 3 genotypes respectively. It was noticed that accessions in cluster-V behaved poorly for the traits under study. Due to poor response, the genotypes designated as drought susceptible and used as testers. Fewer than 100% moisture level, a PC biplot in Figure 4 exhibited that variables and genotypes are super imposed on the plot as vectors. The distance of variables with respect to PC-1 and PC-2 showed contribution of these variables in the variation of different accessions under study. The biplot exhibited that dry root weight and root length as a whole contributed maximum towards variability in cotton genotypes. Fewer than 70% moisture level, a PC biplot in Figure 5, exhibited that variables and genotypes are super imposed on the plot as vectors. The distance of variables with respect to PC-1 and PC II showed the contribution of these variables in the variation of different accessions under study. It was found that shoot length and dry root weight as a whole contributed maximum towards variability in cotton genotypes under study. Whereas a PC biplot (under 40% moisture level) exhibited that variables and genotypes are super imposed on the plot as vectors (Figure 6). The distance of variables with respect to PC-1 and PC-2 manifested the contribution of these variables in the variation of different accessions under study. The biplot showed that shoot length and root shoot length ratio as a whole contributed maximum towards variability in cotton genotypes.

From the data analyzed for cluster and biplot analysis, five cotton genotypes named BH-176, MPS-11, DPL-45, Tree cotton and Cyto-62 can be declared as drought tolerant as these entries showed constant better performance under controlled and at both stress conditions. On the other hand, GS-444, Cooker-315 and MS-64 fall in susceptible genotypes which showed minimum performance at three moisture levels were identified for breeding program.

Discussion

The present study was conducted on root shoot parameters of cotton genotypes at 45 days seedling stage to assess variability under three water levels. It was noticed that water stress effect was more prominent on shoot as on root traits in the present investigation. Previous researchers as Iqbal et al. [23] and Riaz et al. [14] have conducted trials for seedling parameters for germplasm screening to identify drought tolerant and susceptible accessions. In this study, 50 cotton genotypes were checked at seedling stage to determine the variability in nine morphological traits i.e., root length, fresh root weight, dry root weight; root: shoot length ratio, fresh root: shoot weight, ratio, dry root: shoot weight ratio, shoot length, fresh shoot weight, dry shoot weight at controlled and two water stressed conditions. These seedling parameters are adversely affected by moisture stress [21,24]. Means of these traits were compared with cluster and biplot analysis.

Significant reduction was observed in these traits under study. It was found that root growth effected less than shoot growth. Similar findings were given by Govindaraj et al. [23,25] about effect of drought on shoot length of *Pennisetum galucum* and *Gossypium hirsutum L*. while, adverse effect observed by Huang et al. [26] on root growth in tall fescue due to water stress. Observations were given that root associated parameters controlled genetically [1,14,27] but different the case due to environmental factors [28]. The presence of significant variability on genetic basis among 50 cotton accessions under different moisture levels suggested the possible application of such valuable and useful information of seedling traits in cotton breeding program to emerge water stress germplasm [29]. Drought significantly influenced the cotton genotypes performance except BH-176, MPS-11, DPL-45, Tree cotton and Cyto-62 that gave consistence performance for root shoot parameters at different moisture levels [21,30]. Root traits have direct correlation with drought tolerance in rice and cotton crop Nguyen et al. [31] and Khalid et al. [1] respectively. Cluster and biplot technique were applied efficiently to screen germplasm for drought tolerance [32,33]. After the use of statistical approaches for analyzing the data, 5 accession i.e., BH-176, MPS-11, DPL-45, Tree cotton and Cyto-62 found to be water stress tolerant as these entries showed constant better performance under controlled and at both stress conditions. In contrast, GS-444, Cooker-315 and MS-64 fall water stress sensitive genotypes due to minimum performance.

Conclusion

The accessions of upland cotton were diverged from susceptible to tolerant in different groups based on the data on various traits related to water stress conditions. In addition, divergence of the upland cotton give clue that genetic variability is existed within the germplasm and is a valuable source for use in breeding program after another assessment on large area affected with water stress.

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