Comparison of Associations among Yield and Yield Components in Rice (Oryzae sativa L.) under Simulated Drought Stress Condition using Multivariate Statistics

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Abstract — Aim of study was to screen out rice (Oryzae sativa L.) genotypes with better drought tolerance. For exploring observable responses of rice plants towards drought stress, multivariate statistics were used to compare the relationships among the response variables as well as their effects on yield and yield reduction under drought stress. Genotypes responded differently under same level of stress. Results revealed that as a response to drought, all the genotypes showed 42 % average increase in seedling root length; 39 % average decrease in seedling shoot length; 21 % average reduction in 1000 grain weight; delay in heading time of 13 days in average; average yield per plant reduction of 47 %; reduction in grains per panicle of 52% in average, when subjected to simulated drought stress. It was concluded that in addition to correlation and path analysis of yield components, cluster analysis, factor analysis and principal component analysis are also very important methods for screening drought tolerant cultivars of rice in large populations and these methods also give better understandings of associations and helps better to choose important variables and genotypes. Factor analysis and Principal component analysis revealed the importance of drought response index, as a best measurement for exploring genotypes with better drought tolerance. Seedling root-to- shoot length ratio, followed by seedling root length contribute majorly in yield per plant under drought stress. Reduction in plant height and delay in heading time were main causes of yield reduction under water stress. It was further concluded that selecting the genotypes showing higher drought response index, higher increase in seedling root length, seedling root-to-shoot length ratio and less reduction in plant height and delay in heading time, are important criteria in screening for drought tolerance. Also these characters showed high heritability and genetic advance, signifying their values for selection of higher yielding genotypes while higher phenotypic correlation values than their respective genotypic correlation values, indicate their polygenic behavior and high influence of environment.

Index Terms— Oryzae sativa L., Drought stress, Yield reduction, Yield stability, Drought related responses, Correlation analysis, Path analysis, Factor analysis, Principal component analysis, Cluster analysis.

1 Introduction

rought, generally accepted by scientists, is: "a shortfall of water availability sufficient to cause loss in yield", or "a period of no rainfall or irrigation that affects crop growth" (Price [1], Fukai and Cooper [2]). Drought stress is a major constraint to rice (*Oryzae sativa* L.) production and yield stability in rainfed ecosystems (Dey and Upadhyaya [3]). The global reduction in rice production due to drought averages 18 M t annually. In Asia alone, it is estimated that a total of 23 M ha of rice fields (10 M ha in upland and 13 M ha in lowland) are drought-prone (Pandey *et al.* [4]).

Drought is also a particularly important production constraint in Pakistan, with more than 10 M ha of drought-prone fields, where yield losses due to droughts are reported to cost an average of 250 million US\$ per year (Anonymous [5]). So in developing drought resistant cultivars especially with good performance under late season; drought stress is one of the major objectives in rice breeding programs (Boojung and Fukai [6] and Pantuwan *et al.* [7]). However, the progress in

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breeding for drought resistance is rather slow in rice due to complexity of the traits.

There is a need to develop rice varieties that are better yielders under both drought and favorable environments, and are least affected by drought. Many traits like grains per panicle yield per plant, heading time and drought response index are known to contribute in improving yield under drought, but their actual contribution towards yield reduction in not understood. Also comparing the responses of rice genotypes, in terms of change, increase or decrease in characters due to stress is better criteria to screen out drought tolerant varieties, than comparing the characters only in drought stress. Therefore, increase and decrease in characters were measured and compared with one another as well as with reduction in yield as resultant variable in order to find out actual parameters that majorly contribute in yield reductions under drought stress.

Genotypic and phenotypic correlation coefficient analysis is generally used to measure the relationship between two traits (Kown and Torrie [8]). But it gives not enough information about the relationships between different traits. Therefore, other statistical methods such as cluster analysis, factor analysis and principal component analysis were also included to interpret the results. These multivariate statistical analyses can provide more insights on the deep structure of data and traits' relationship (Bramel *et al.* [9] and Allard [10]).

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2 MATERIAL AND METHODS

2.1 Experimental procedures:

Two seeds of each variety were sown into one pot. Distance between seedlings was 16.5 cm, within each pot. The soil used for planting was collected from same source, air dried and mixed thoroughly. The experiment was conducted in two conditions: full water condition (control) and water limited condition creating a simulated drought stress. The moisture contents were maintained by using moisture meter (T-NH2, Cambridge, England) by applying water on alternate days in specific volume. For creating simulated drought stress condition, two consecutive drying cycles were imposed so that plants might not be fully dried. First dry cycle was imposed at reproductive stage while the next one was imposed when plants started panicle emergence. Drought stress was removed by irrigating the pots to full water capacity. Both of the treatments were replicated three times under a completely randomized design. Phosphorus (P) and Potassium (K) were applied in full dose at the time of sowing; Nitrogen (N) was applied in four splits as top dressing. Insect and weed control measures were applied as normal. Traits were measured in both the conditions and their differences were calculated to measure the response of genotypes towards drought stress.

Drought tolerance indexes e.g. increase in seedling root length (inc. RL), decrease in seedling shoot length (dec. SL), increase in seedling root shoot length ratio (inc. R/S), drought response index (DRI), yield reduction (YR), reduction in number of grains per panicle (dec. G/P), decrease in 1000 grain weight (dec. TGW), reduction in plant height (RPH) and delay in heading time (DHT) were recorded. The following genotypes were used for conducting experiment collected from Kala Shah Kakoo Rice Research Institute, Lahore Pakistan, SRS504, SRS54, SRS65, SRI52, SRI31, SRI61, SRI 1-8, China1, SRS502, SRS503, SRS62, SRS64, SRI57, SRS63, SRI53, SRI 51, IRRI-6, China2, SRI 16.

2.1.1 Seedling root and shoot length measurements:

Root traits were measured at seedling stage of each line growing in polythene bags, two seeds per bag, under both well water and simulated drought stress conditions. The experiment was carried out in two sets (one for normal and the second for drought stress condition) with three replications following a completely randomized design. Drought stress was applied to one set while the other one was used as control. For measuring root length, the seedlings were pulled out of soil carefully without damaging the roots. Then the roots were clearly washed with distilled water and laid on a stand provided with a scale. The longest visible root from the basal node was scored as the seedling root length (cm). Likewise, the length of shoots was measured from the basal node to the longest shoot (cm).

2.1.2 Delay in heading time (DHT):

Delay in heading time was calculated as the differences of heading date under drought stress and control conditions. Heading date was determined visually when about 50% of the tillers in each pot were headed. The days to heading of each

genotype were counted from day of sowing of nursery up to the day of emergence of panicle.

2.1.3 Reduction in plant height (RPH):

Reduction in plant height was calculated as the differences of plant heights under drought stress and control conditions. Plant heights were measured from ground level to the tip of the plant using a meter rod when all of the tillers in each pot were headed. These measurements were then averaged.

2.1.4 Yield per Plant and Drought Response Index:

The plants under normal and stress conditions were separately harvested to measure the grains yields and DRI was calculated by the following formula given by Song-ping [11]:

DRI = the grain yield under stress / the grain yield under normal condition

2.1.5 Reduction in Number of grains per panicle:

Number of grains/panicle was counted for each genotype. Difference in number of grains per panicle was calculated as the differences of grains per panicle under drought stress and control conditions.

2.1.6 Measurement for Thousand Grain Weight:

The seeds from all the plants in a replication of a genotype were bulked and thousand grains were counted and weights were recorded in grams. The reduction rate in 1000 grain weight was calculated as the differences of 1000 grain weight under drought stress and control conditions.

2.1.7 Reduction in Yield per plant:

Total grains obtained from each plant in a replication were weighed in grams. Then the measurements were averaged for yield per plant for each genotype. The reduction in grain yield per plant was calculated as the differences of total grain weight per plant under drought stress and control conditions.

2.2 Statistical Analyses:

2.2.1 Analysis of variance:

ANOVA (Steel *et al.* [12]) was performed for response of each trait i.e. statistical differences in that trait as measured in both normal and simulated drought stress condition.

2.2.2 Broad Sense Heritability and Genetic Advance:

Heritability in broad sense was estimated according to Falconer and Mackay [13]; Genetic advance was computed at 10 % selection intensity (i = 1.755) using formula given by Poehlman and Sleper [14].

Broad sense heritability = V_g/V_p Genetic advance = $\sqrt{V_p} \times h_{b.s}^2 \times 1.755$

2.2.3 Correlation Coefficient Analysis:

Genotypic and phenotypic correlation coefficients of all the traits under study were estimated according to the statistical procedure as described by Kown and Torrie [8]. Value of genotypic correlation was considered significant if its absolute

value was greater than twice of its standard error. Likewise, a phenotypic as well as environmental correlation coefficient was considered significant if t-calculated was greater than t-tabulated.

2.2.4 Path coefficient analysis:

Path coefficient analysis was performed according to method given by Dewey and Lu [15], using genotypic correlation coefficients. Yield per plant was kept as resultant variables (effect) and the responses of plants as casual variables (causes).

2.2.5 Cluster Analysis:

Cluster analysis (Eisen *et al.* [16]) was applied in order to arrange the cultivars/genotypes in various groups and subgroups to find the genotypes that showed similar behavior under drought stress and to find the outliers among the studied genotypes. It was also used to find out the distant genotypes to be used in breeding program and hybrid production for heterosis. Cluster analysis was also performed for variables for arranging the studied traits into different clusters in order to compare the traits that are more similar and correlated to one another within and among the clusters.

2.2.6 Factor Analysis:

Factor analysis consists of lessening of a large number of associated variables to a much smaller number of uncorrelated variables. Main factor is found out and then the matrix of factor loading was used to varimax rotation, and the communality and variances of uncorrelated variables was estimated by highest correlation coefficient in each array as suggested by Seiller and Stafford [17].

2.2.7 Principal Components Analysis:

PC analysis (Bramel *et al.*[9]) is a mathematical procedure performed to classify a large number of variables (genotypes and traits) into major components and calculate their contribution to the total variation. The first PC is the variable with highest variability among all the traits followed by the second one.

3 RESULTS:

3.1 Analysis of Variance:

ANOVA showed that all the genotypes responded differently under simulated drought stress, enlightening presence of variation in genotypes for all studied characters. As a response to drought, all the genotypes showed 42 % average increase in seedling root length ranging from 27%-64%; 39 % average decrease in seedling shoot length ranging from 3%-34%; 21 % average reduction in 1000 grain weight ranging 5-39%; delay in heading time of 13 days in average ranging 8-23 days; average yield per plant reduction of 47 % ranging from 21-84 %; reduction in grains per panicle of 52% in average ranging from 9-94 % when subjected to simulated drought stress (Table 2).

3.2 Variances and coefficient of variability:

Phenotypic coefficients of variability (PCV) of almost all the traits were much higher than their respective genotypic coefficients of variability (GCV), except decrease in grains per

panicle and decrease in yield per plant, foe which PCV values are higher but nearly equal to their respective GCV values (Table 3).

3.3 Broad sense heritability and Genetic Advance:

Decrease in yield per plant and decrease in grains per panicle showed maximum Broad sense heritability (0.997 and 0.998 respectively) and Genetic Advance (27.672 and 51.848 respectively). Higher heritability estimates for increase in root length (0.878), increase in root-to-shoot length ratio (0.871) and delay in heading time (0.938) were also observed (Table 3).

Results show that all the genotypes showed significant difference in yield from one another under normal as well as simulated drought stress condition (Table 2). The mean grain yield was 11.42 g/plant under simulated drought stress conditions while mean grain yield per plant under normal condition was recorded to be 23 g / plant. Yield maintenance percentage was recorded equal to 53 % as a whole in average (table 2). The results also showed that the genotypes that showed more yield under normal condition also performed better under simulated drought stress condition. A few lines had high yield under both conditions (figure 1), but most of the lines had high yield only under favorable condition and did not perform well under drought stress conditions (Zou *et al.* [18]).

Score plot (figure 1) shows scoring of genotypes against yield stability (decrease in yield per plant under drought stress) and yield potential under drought (yield per plant under stress) and their performances under drought stress condition relative to these factors. The genotypes at right to the vertical reference line are higher yielders under drought stress while on left are lower yielder. The genotypes near the vertical reference line are showing average performance. Likewise, those genotypes that are below the horizontal reference line are greatly affected by the stress and showed higher reduction in yield as compared to those above the reference line. So the plot area may be divided into four partitions. The cultivars that are in square (b) within the circle, may be used in breeding program for drought stress, because these genotypes showed higher yield in stress environment and also showed less reduction in yield due to stress. These may also be used in back cross breeding or hybridization program. SRS 503 was best genotype in terms of yield and yield stability under drought stress, followed by SRI 57. While china 1 was least yielder and least stable genotype among all the genotypes studied followed by SRI 61. China 2 was a good yielder under normal condition and performed better in full water condition, but highly affected by drought stress and showed abrupt reduction in yield on experiencing drought.

3.4 Correlation analysis:

3.4.1 Yield per plant:

Genotypic correlation coefficient for yield per plant under drought condition was positive and significant with increase in seedling root length, highly significant with increase in seedling root to shoot length ratio, drought response index; while negative and significant with decrease in 1000 grain weight and highly significant with delay in heading time, decrease in yield per plant and reduction in plant height due to drought stress. Almost same results were observed at phenotypic level (Table 4). Same results were detected by other researchers in different experiments (Pandey *et al.*[4], Pantuwan *et al.*[7], Surek and Baser [19a], Surek and Baser [19b], Raju *et al.* [20], Price *et al.*[21], O'Toole [22] and Kanbar and Shashidhar [23]).

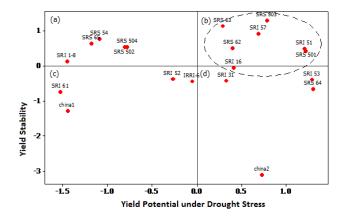


Fig 1: Score plot of all genotypes against yield per plant (yield potential) and decrease in yield (yield stability) under simulated drought stress condition

3.4.2 Seedling characters:

Increase in seedling root length, at genotypic level, was highly significant and positively correlated with increase in seedling root to shoot length ratio (0.870), yield per plant (0.350) and drought response index (0.361); and significant positive with decrease in seedling shoot length (0.277). While it showed negative and significant negative relation with reduction in plant height (-0.457) and decrease in yield per plant (-0.327). Likewise, at genotypic level, increase in seedling root to shoot length ratio showed highly significant and positive relation with yield per plant (0.238) while negative with reduction in plant height (-0.325). Seedling shoot length was highly negative correlated with decrease in thousand grain weight (-0.545) and decrease in yield per plant (-0.541) and DRI (-0.486).

3.4.3 Delay in heading time:

Drought stress that developed prior to flowering generally delayed the time of flowering of genotypes, and the delay in flowering was positive and significantly correlated with decrease in 1000-grain weight (0.301), decrease in yield per plant (0.561) and reduction in plant height (0.771) under simulated drought stress condition while negatively correlated with yield per plant (-0.599) under drought stress and drought response index.

3.4.4 Reduction in plant height:

Genotypic correlation of reduction in plant height under drought condition (Table 4) has positive and significant correlation with delay in heading time (0.771) and decrease in yield per plant (0.766) under simulated drought stress condition while negatively correlated with drought response index (-0.794) and yield per plant (-0.740) under drought stress [24]. At phenotypic level, same results were observed.

3.4.5 Decrease in Thousand Grain Weight:

Decrease in thousand grain weight was positively associated with delay in heading time (0.301), decrease in grains per panicle (0.678) and reduction in plant height (0.742) while negatively correlated with decrease in seedling shoot length (-0.545), increase in seedling root length to shoot length ratio (-0.790), and yield per plant (-0.575) under drought stress.

3.5 Path Analysis:

The correlation coefficients were partitioned into direct and indirect effects (Tables 5). Yield per plant under drought stress was taken as resultant variable while all the other traits as causes. Reduction in yield per plant (-0.767) showed highest but negative direct effect on yield per plant under stress; followed by decrease in thousand grain weight (-0.657). Among the other characters, increase in seedling root to shoot length ratio had maximum direct effect (-0.310) followed by DRI (0.308), while direct effects of the other variables were relatively low.

3.6 Factor Analysis:

Data presented in table 1 and figure 2 clearly demonstrates that the increase in number of components was negatively associated with eigenvalues. Figure 2 clearly shows that first two factors, out of all the factors, accounted for 65.9 % of the total variations (Table 1). The first factor was included for DRI, reduction in plant height, decrease in yield per plant, yield per plant under drought stress and delay in heading time and accounted for 37.9% of the total variation; while the second factor was included for increase in seedling root to shoot length ratio, increase in seedling root length and decrease in shoot length that accounted for 26% of the total variation. Rotated Factor Loadings and Communalities (Table 1b) clearly shows that the first factor includes all the traits that are negatively associated with yield under stress and also the reduction in yield is positively correlated; while yield per plant under stress is negatively correlated with the first factor, so it can be named as yield reducing factor under stress. On the other hand, second factor can be named as yield mantaining factor, because traits (increase in seedling root length, increase in seedling root to shoot length ratio and decrease in seedling shoot length) including yield per plant under stress is negatively associated with second factor. Both the factors are graphically depicted in figure 3 and 4. So the genotypes can also devided into two clusters. Cluster I comprises of the better performing genotypes, while cluster II contains genotypes that performed worst in drought prone environment (Fig 3). Variable loadings by factor analysis and varimax roration with first two factors under simulated drought stress condition is depicted in figure 4 and table 1 respectively. In context of figure 3, SRI 52 is located in lowest of second factor, indicating that it shows highest increase in its seedling root length and seedling root to shoot length ratio, while IRRI-6 showed the lowest. Similarly, SRI 1-8 has highest first factor and showed most reduction in plant height, delayest in heading date and most reduction in yield among

all the genotypes.

TABLE 1 FACTOR ANALYSIS OF THE CORRELATION MATRIX

(A) Unrotated Factor Loadings and Communality	(A)	Unrotated	Factor	Loadings	and	Communalit	ies
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Variable	Factor1	Factor2	Communality
y/p d	-0.876	-0.095	0.777
Dec. y/p	0.832	-0.433	0.880
Inc. RL	-0.492	-0.556	0.551
Dec. SL	0.067	-0.844	0.717
Inc. R/S	-0.259	-0.823	0.744
Dec. TGW	0.455	0.358	0.519
DHT	0.693	-0.218	0.528
Dec. Grains/p	0.309	0.398	0.254
RPH	0.860	-0.021	0.741
DRI	-0.832	0.433	0.880
Variance	3.9891	2.6016	6.5907
% Var	0.399	0.260	0.659

(B) Rotated Factor Loadings and Communalities (Varimax Rotation)

Variable y/p d Dec. y/p Inc. RL Dec. SL Inc. R/S Dec. TGW DHT Dec. Grains/p RPH DRI	Factor1 -0.773 0.935 -0.243 0.384 0.075 0.207 0.724 0.134 0.803 -0.935	Factor2 -0.423 0.083 -0.702 -0.754 -0.859 0.690 0.063 0.486 0.309 0.083	Communality 0.777 0.880 0.551 0.717 0.744 0.519 0.528 0.254 0.741 0.880	
Variance	3.7869	2.8038	6.5907	
% Var	0.379	0.280	0.659	

3.7 Principal Component Analysis:

Table 6 shows that 79.6% of the variations are explained by the first three principal components. Of which 65.9% variation is due to first two components. Out of this variability, 39.9% and 26% of the total variability is explained by first and second component respectively (Table 6). PC1 was moderately positively correlated with decrease in yield per plant (0.417), reduction in plant height (0.431), delay in heading time (0.347); while it showed negative correlation with yield per plant under stress (-0.439) and drought response index (-0.417). PC2 was highly negative correlated with decrease in seedling shoot length (-0.523), increase in seedling root to shoot length ratio (-0.510), increase in seedling root length (-0.345) and positively with decrease in thousand grain weight (0.346) under drought stress. So, the estimated drought related rice responses were grouped into two components, yield reducing (PC1) and yield mantaining (PC2) and their contribution in the variables for drought stress are graphically represented in figure 5. Both the components and their contribution in the traits for simulated drought stress condition are graphically presented in fugure 5.

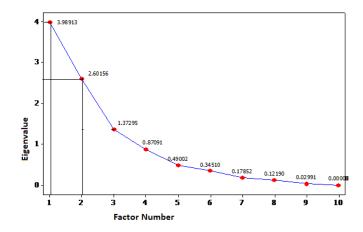


Fig 2: Scree plot showing eigenvalues in response to number of factors for the estimated variables of rice under simulated drought stress condition

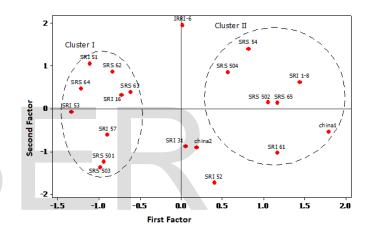


Fig 3: Score plot of all genotypes on first and second factors under drought stress

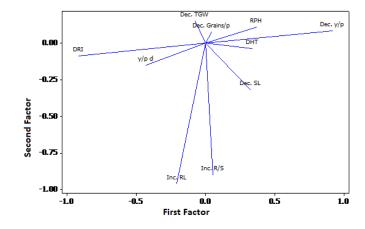


Fig 4: Variable loading by factor analysis and varimax roration with first two factors under simulated drought stress condition.

Table 2

Mean Square Values for some Drought Related Traits under Simulated Drought Stress Condition

Characters	Inc. RL	Dec. SL	Inc. RL/SL	Dec. TGW	DHT	Dec. Y/P	Dec. G/p	RPH
Treatment SS	764.083**	114.345*	13.160**	615.072*	1113.645**	22427.916**	78488.480**	83.632*
Replication SS	18.463	22.033	6.724	48.902	435.600	230.400	448.900	336.400
Error SS	35.227	68.633	1.236	36.108	48.400	45.600	89.100	59.600
Total SS	817.774	205.012	21.120	700.082	1597.645	22703.916	79026.480	479.632
G.M.	9.762(cm)	3.029(cm)	1.109	6.020 (g)	2.755 (Days)	46.861 (g)	5.857	9.825(cm)
Replication MS (df = 2)	9.232	1.017	3.362	24.451	217.800	115.200	224.450	168.200
<pre>Treatment MS (df = 19) MS1</pre>	38.636	6.018	0.693	21.846	58.613	1180.417	4130.973	4.402
Error MS2	1.717	2.332	0.033	3.213	1.274	1.200	2.345	1.568

Table 3

ESTIMATES OF GENOTYPIC, PHENOTYPIC AND ENVIRONMENTAL VARIANCES AND COEFFICIENT OF VARIABILITY FOR DROUGHT RELATED TRAITS

UNDER SIMULATED DROUGHT STRESS CONDITION

Characters	Inc. RL	Dec. SL	Inc. RL/SL	Dec. TGW	DHT	Dec. Y/P	Dec. G/p	RPH
Genotypic variances (Vg)	12.306	1.229	0.220	5.211	19.113	393.072	1376.209	0.944
Phenotypic variances (Vp)	14.023	3.561	0.253	11.424	20.387	394.272	1378.554	2.513
Environmental variances (Ve)	1.717	2.332	0.033	6.213	1.274	1.200	2.345	1.568
Phenotypic coefficient of Variability (PCV)	38.361	62.307	45.316	56.143	35.399	42.373	633.959	16.134
Genotypic coefficient of Variability (GCV)	35.936	36.597	42.298	37.917	34.276	42.308	633.420	9.891
Heritability (h2 B.S.)	0.878	0.345	0.871	0.456	0.938	0.997	0.998	0.376
GA	4.310	0.535	0.572	1.458	5.738	27.672	51.848	0.511

Inc. RL=increase in root length, dec. SL=decrease in shoot length, inc. RL/SL=increase in root length to shoot length ratio, dec. TGW=decrease in thousand grain weight, dec.G/P=decrease in grains per panicle, dec. Y/P=decrease in yield per plant, Y/P*=yield per plant under drought stress, RPH=reduction in plant height, DHT=delay in heading time, DRI=Drought Response Index

Table 4

ESTIMATES OF GENOTYPIC CORRELATION (ABOVE) AND PHENOTYPIC CORRELATION (BELOW) COEFFICIENTS FOR DIFFERENT DROUGHT RELATED TRAITS

UNDER SIMULATED DROUGHT STRESS CONDITION

Characters	Dec. SL	Inc. RL/SL	Dec. TGW	DHT	Dec. G/P	RPH	Dec. Y/P	DRI	Y/P
Inc. DI	0.277*	0.870**	-0.247ns	-0.121ns	-0.163*	-0.457*	-0.327**	0.361**	0.350**
inc. RL	0.195*	0.754**	-0.241ns	-0.136ns	-0.150*	-0.139ns	-0.298*	0.281*	0.081*
og GT		0.790**	-0.545**	0.142*	-0.177ns	-0.193ns	0.541**	-0.486*	0.060ns
ec. SL		0.555**	-0.310*	0.066ns	-0.117ns	0.105ns	0.319**	-0.14 ns	0.065ns
DI /GI			-0.325*	0.078*	-0.181ns	-0.234ns	0.035ns	-0.019ns	0.238**
nc. RL/SL			-0.289*	0.067*	-0.174ns	-0.040 ns	0.026ns	-0.041*	0.216**
o a more				0.301**	0.678*	0.742**	0.010ns	-0.139ns	-0.575**
ec. TGW				0.226*	0.468*	0.114ns	0.005ns	0.093ns	-0.338*
· · · ·					0.077ns	0.771**	0.561**	-0.599**	-0.491**
HT					0.078ns	0.477**	0.542**	-0.508**	-0.455**
C/D						0.117ns	0.072*	-0.094ns	-0.331*
ec. G/P						0.065ns	0.072*	-0.079ns	-0.316*
DII							0.766**	-0.794**	-0.740**
PH							0.475**	-0.469**	-0.607**
og V/D								-0.849*	-0.634**
Dec. Y/P								-0.952**	-0.615**
DT									0.642*
RI									0.628**

^{*} Significant at 5 % confidence level, ** Significant at 1 % confidence level

Table 5

ESTIMATES OF DIRECT (BOLD FIGURES) AND INDIRECT EFFECTS (VERTICALLY ARRANGED) OF DIFFERENT DROUGHT RELATED TRAITS ON YIELD UNDER SIMULATED DROUGHT STRESS CONDITION

Characters	Inc. RL	Dec. SL	Inc. RL/SL	Dec. TGW	DHT	Dec. G/P	RPH	Dec. Y/P	DRI
Inc. RL	-0.310	-0.086	-0.27	0.077	0.038	0.051	0.142	0.102	-0.112
Dec. SL	0.065	0.236	0.187	-0.129	0.033	-0.418	-0.045	0.128	-0.115
Inc. RL/SL	0.159	0.145	0.184	-0.059	0.014	-0.033	-0.042	0.006	-0.003
Dec. TGW	0.162	0.358	0.213	-0.657	-0.197	-0.446	-0.488	-0.007	0.091
DHT	0.019	0.024	0.012	0.048	0.158	0.012	0.121	0.009	-0.095
Dec. G/P	0.033	-0.031	-0.037	0.136	0.015	0.202	0.023	0.015	-0.019
RPH	0.037	-0.016	-0.019	0.061	0.063	0.009	0.081	0.062	-0.065
Dec. Y/P	0.251	-0.415	-0.027	-0.008	-0.430	-0.055	-0.588	-0.767	0.652
DRI	0.111	-0.149	-0.006	-0.043	-0.184	-0.029	-0.244	-0.261	0.308

Residual effect = **0.15**

Inc. RL=increase in root length, dec. SL=decrease in shoot length, inc. RL/SL=increase in root length to shoot length ratio, dec. TGW=decrease in thousand grain weight, dec.G/P=decrease in grains per panicle, dec. Y/P=decrease in yield per plant, Y/P*=yield per plant under drought stress, RPH=reduction in plant height, DHT=delay in heading time, DRI=Drought Response Index

TABLE 6
EIGENVALUES AND THE CORRELATION MATRIX FOR THE ESTIMATED VARIABLES OF RICE USING PRINCIPAL COMPONENT PROCEDURE FOR DROUGHT TOLERANT CULTIVARS UNDER DROUGHT STRESS CONDITION

Variable	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10
y/p d	-0.439	-0.059	-0.114	0.073	-0.528	-0.051	0.238	0.658	-0.120	-0.000
Dec.y/p	0.417	-0.269	-0.188	0.190	-0.004	0.218	0.355	0.074	-0.062	0.707
Inc. RL	-0.246	-0.345	0.481	-0.230	0.228	0.371	0.083	-0.011	-0.582	0.000
Dec. SL	0.034	-0.523	0.001	0.396	-0.090	-0.589	-0.314	-0.129	-0.313	0.000
Inc. R/S	-0.130	-0.510	0.408	-0.045	0.078	-0.078	0.164	0.071	0.714	-0.000
Dec. TGW	0.228	0.346	0.497	-0.017	-0.114	-0.481	0.553	-0.086	-0.155	0.000
DHT	0.347	-0.135	0.178	-0.398	-0.735	0.159	-0.268	-0.183	0.017	0.000
Dec. Grains/p	0.155	0.247	0.485	0.643	-0.074	0.321	-0.318	0.230	0.061	-0.000
RPH	0.431	-0.013	0.065	-0.371	0.311	-0.228	-0.285	0.664	-0.051	-0.000
DRI	-0.417	0.269	0.188	-0.190	0.004	-0.218	-0.355	-0.074	0.062	0.707
Eigenvalue	3.9891	2.6016	1.3730	0.8709	0.4900	0.3451	0.1785	0.1219	0.0299	-0.0000
Proportion Cumulative	0.399	0.260 0.659	0.137 0.796	0.087 0.883	0.049	0.035 0.967	0.018 0.985	0.012 0.997	0.003 1.000	-0.000 1.000

Inc. RL=increase in root length, dec. SL=decrease in shoot length, inc. RL/SL=increase in root length to shoot length ratio, dec. TGW=decrease in thousand grain weight, dec.G/P=decrease in grains per panicle, dec. Y/P=decrease in yield per plant, Y/P*=yield per plant under drought stress, RPH=reduction in plant height, DHT=delay in heading time, DRI=Drought Response Index

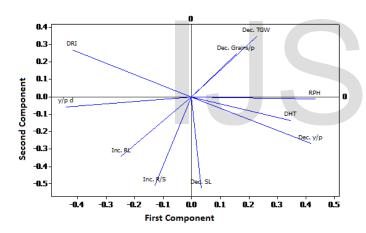


Fig 5: Loading plot of variables with first two components under simulated drought stress condition

3.8 Cluster Analysis:

Hierarchical cluster (Everitt and Dunn [47]) analysis among the genotypes showed that there was similarity ranging from 59% to 79% under simulated drought stress condition. Studied genotypes could be grouped into two major and two minor clusters, on the basis of their performances under drought stress (figure 6). Genotypes lying in third cluster were best performing under drought stress and more stable. Almost all the genotypes in this group also had high DRI, high seedling root to shoot length ratio, showed higher TGW and less reduction in plant height and delay in heading time when subjected to drought stress. Likewise the drought related traits/variables of rice genotypes could be agglomerated into four clusters (figure 7). Dendrograms based on different linkage methods (Single and average) with simple correlation co-

efficients are shown in Figure 7a and b. Since different linkage methods are giving same four clusters, it is easier to interpret the associations among traits.

4 DISCUSSION

The results of analysis of variance indicated that the genotypes responded differently in terms of all studied traits under stress condition. The results (table 3) also showed that the phenotypic coefficients of variability of all the genotypes were much higher than its respective genotypic coefficient of variability for all traits, indicating the environmental influences that constitute the major portion of the total phenotypic variation and the traits are controlled by poly-genes (Mahto et al.[25], Habib et al.[26], Satyanarayana et al.[27]. Higher heritability estimates for decrease in grains per panicle, followed by decrease in yield per plant, delay in heading time, increase in root length and increase in root to shoot length ratio due to stress combined with high genetic advance indicated the presence of additive genes (Balan et al.[28]). Higher heritability estimates also indicate that these characters can be exploited more efficiently through selection in further generations. Same results were also concluded by other researchers (Girish et al.[24], Qamat et al.[29], Baber et al.[30], Okelala et al.[31], Zahid *et al.*[32] and Prasad *et al.*[33]).

Correlation coefficients help to measure the extent and direction of association between two variables. Negative and highly significant relationship of yield per plant with decrease in yield per plant indicates that the genotypes that are high yielder under well watered condition may also be the high yielder under drought stress (Habib *et al.*[26], Iftikharuddaula *et al.*[34], Hsiao [35], Hasib [36], Dang *et al.*[37], Ali *et al.*[38] and Ali *et al.*[39]). Increase in seedling root

length showed significant and positive relationship with decrease in seedling shoot length, indicating shoot length was lessened as root length was increased under drought stress. It further elucidates that plants tolerate drought at seedling stage by lowering their shoot development at earlier stages and utilize this energy in developing and increasing root system. More efficient root system developed by better performing plant under drought stress may be advantageous for extracting more water from drying soils, which explains its better growth under stressed conditions (Pantuwan et al. [7], Dang et al.[37], Yue et al.[40] and Yang et al.[41]). The results further clarify the fact that unavailability of water and drought stress stimulates root growth in order to explore more water for its normal development (Chopra and Paroda [42]). The genotype IRRI 6 and china 1 showed minimum increase in root length under drought stress and could not show better performance and showed maximum yield loss under drought prone environment, while SRS 503, SRS 501 and SRI 57 showed maximum increase in root depth under simulated drought stress condition and also showed minimum yield loss due to stress.

Moreover, decrease in seedling shoot length was also positively and highly significant with decrease in yield per plant. Table 2 clearly emphasizes that different genotypes showed pronouncedly different rate of seedling shoot diminution at seedling stage when subjected to same level of drought stress. The finding also coincides with the finding of Kanbar and Shashidhar [23].

Drought stress that developed prior to flowering generally delayed the time of flowering of genotypes. Genotypes with a longer delay in flowering time had extracted more water during the early drought period, and as a consequence, had higher water deficits and less yielder (Pantuwan et al. [7]). When drought is developed during late growth stages and occurs for a short period, early to escape water stress is normally an important character. But in most of the dry areas, this is not the same case and drought mostly occurs for longer durations. So, when drought was implemented for longer duration, genotypes that delayed flowering experienced more drought stress and hence showed more decrease in thousand grain weight, less grains per panicle resulting in less yield. In the present study, it may be concluded that those genotypes that showed less delay in heading time due to stress, performed better in drought stress as compared to those lines that showed more delay in heading date due to stress. In short, drought resistant lines had a shorter delay in heading date than susceptible lines (Sieller and Stafford [17]). This finding that drought stress lengthened the duration of the heading date is consistent with the other experiments, in which drought was developed near the heading date (Pantuwan et al. [7], Lilly and Fukai [43], Boojung and Fukai [44] and Wopereis [45]). Under drought stress condition, genotypes with a longer delay in flowering were further disadvantaged because they experienced a larger water deficit at flowering when the soil moisture decreased with time.

Results also indicated that the genotypes with a larger reduction in plant height, also showed a larger reduction in yield due to drought stress, fewer yields and less grain per

panicle under drought stress (Price [1], Sieller and Stafford [17] and Babu *et al.* [46]). The reduction in plant height may be associated with internal plant water status, particularly turgor pressure to exert the panicle and Internal plant water deficit inhibiting cell expansion and growth of plant (Hsiao [35]).

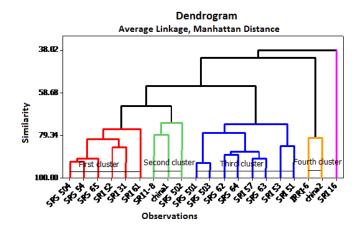
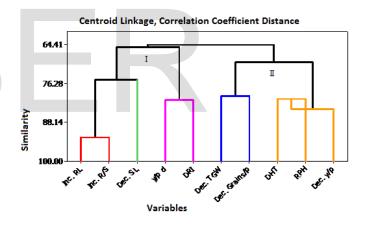


Fig 6: grouping of 20 genotypes for drought related traits on basis of average linkage under Manhattan distance



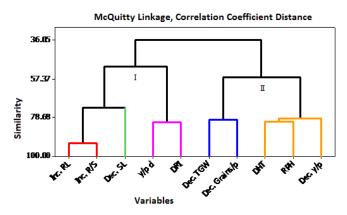


Fig 7: Dendrograms showing similarity levels on basis of average, centroid and McQuitty linkage with simple correlation coefficient distance of the estimated ten drought related traits/variables in rice using the hierarchical cluster analysis under simulated drought stress condition

Factor analysis revealed the importance of drought response index, as a best measurement for screening out drought tolerant genotypes. Seedling root length to shoot length ratio, followed by seedling root length contrubuted majorly in yield per plant under drought stress. Reduction in plant height and delay in heading time were main causes of yield reduction under stress (figure 4). Score plot of genotypes further explains the behavior of genotypes individually on the basis of first two major factors. It can be observed that genotypes present in negative side of both the factors were higher yielder and first factor contributed more in reducing the yield under stress as compared to second factor respectively. Cluster II contains the genotypes that were less yielder and showed drstic reduction in their yields due to stress, showed less DRI, more reduction in plant height and were delayer in heading date when subjected to drought stress. Cluster I contains genotypes that showed more yield under stress, less reduction in yield per plant due to stress, high DRI, less reduction in plant height and less delay in heading time. Within the cluster I, genotypes can further be divided into two groups in accordance with second factor, above 0 of Y-axis and below. All genotypes that are below the 0 line showed good yield in terms of better root system and less due to other characters while above it are those genotypes that gave good yield due to other characters and less due to root traits. SRS-503 showed its yield more due to efficient root system, followed by SRS-501.

By the facts that both the factors were negatively correlated with yield per plant under drought, and only the first factor was positivly correlated with decrease in yield per plant due to stress (Table 1 b), it can be anticipated that those characters that are negatively correlated with first factor e.g. DRI, are positively correlated with yield per plant under drought stress while negatively with decrease in yield per plant. Likewise, those characters that are positively correlated with first factor e.g. RPH and DHT, are also positively correlated with decrease in yield per plant while negatively with yield per plant under drought stress (Table 1b). in the same way, those characters that are negatively correlated with second factor e.g. Inc. RL, Dec. SL and Inc. R/S, are positively correlated with yield per plant under drought stress and negative with decrease in yield per plant due to drought stress.

Results of principal component analysis demonstrated the same results with further details. Loading plot of variables with first two components showed that DRI and increase in root length is heavily projected over and nearer to yield per plant under drought stress, followed by increase in seedling root length to shoot length ratio, indicating their levels of positive contribution towards yield per plant under drought condition (Figure 4). On the other hand, reduction in plant height is lying opposite to yield per plant under drought stress showing highest projection over PC1 followed by decrease in yield per plant, dealy in heading time and decrease in thousand grain weight and grains per panicle, indicating their levels of negative association with yield per plant under drought and their role in yield reduction

[47]. Yield per plant under stress and DRI are negatively correlated with PC1 while decrease in yield per plant, delay in heading time and reduction in plant height are positively correlated (Table 6). It indicates that yield per plant under drought stress has relationship with DRI while RPH, DHT and Dec. Y/p showed negative relationship with yield per plant under stress and showed positive relationship with decrease in yield per plant. Likewise, decrease in yield per plant. Inc. RL, Dec. SL and Inc. R/S showed negative correlation with PC2 while Dec. TGW showed positive.

Mode of associations of all the responses with yield per plant under stress and decrease in yield per plant due to stress as well as associations among the responses can be observed clearly from dendrograms in figure 7 (a & b). Cluster analysis (Everitt [48]) among drought related responses showed that yield per plant was closely associated with drought response index, and comparatively less correlated with increase in seedling root length, increase in seedling root length to shoot length ratio while decrease in yield per plant was mainly due to delay in heading time and reduction in plant height. As important characters, positively affecting rice yield per plant under drought stress, DRI, increase in seedling root length, increase in seedling root length to shoot length ratio are in same cluster, lying with yield per plant under drought as shown in figure 7a&b, while traits e.g. delay in heading time, reduction in plant height, decrease in thousand grain weight and decrease in grains per panicle, closely related to decrease in yield per plant, are lying in same cluster of dendrograms.

5 CONCLUSION

The results clarify that those genotypes that delayed their heading time, experienced more drought stress and showed more reduction in plant height, performed poor under drought stress. So, while screening the rice cultivars for the areas that experience longer periods of drought, it will be wiser to select those genotypes that have higher drought response index, possess more ability to extend their root system, show less reduction in plant height and less delay in flowering due to stress.

It was further concluded that simple correlation analysis could only determine the linear relationship between two related variables, but was unable to clearly show how the multiple variables are related to one another depending to resultant variable (yield). Path analysis uses results from simple correlation, so it cannot be used as ultimate analysis to distinguish important traits under complicated condition such as drought, along with cluster analysis. It may be concluded that, under such conditions, principal component and factor analysis are also stronger and helpful methods to be applied for screening important drought related traits and drought tolerant genotypes, and should also be included in interpreting the results in such situations.

6 ACKNOWLEDGMENT

I am extremely grateful to Mr. Abdul Razaq, Mr. Asrar Mehboob, Mr. Saleem ur Rehman and Mr. Aamir Hussain from Maize and Millets Research Institute, Yusufwala, Sahiwal, Pakistan as well as Mr. Aftab Iqbal from Potato Research Institute, Sahiwal, Pakistan, for their paramount support in interpreting results and sharing their essential knowledge for writing discussions. I am also very thankful to Mr. Khalid Mehmood for his appreciative help in analysing data.

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