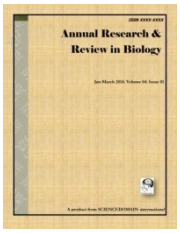

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Genetic Advance Prediction and Multivariate Analysis for Antioxidants and Agronomic Traits in Wheat

Lalehzar Ghaed-Rahimi[1] and Bahram Heidari[1*]

[1]Department of Crop Production and Plant Breeding, College of Agriculture, 7144165186, Shiraz University, Shiraz, Iran.

Authors' contributions

Author LGR conducted experimental works and wrote the first draft of manuscript. Author BH managed the proposal of the study and supervised experimental work and revised the manuscript.

Original Research Article

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ABSTRACT

Background: The Interrelationship of traits is important for structuring crop populations and modeling selection criteria for increasing grain yield.
Aims: Assessing interrelationship of traits under drought stress and normal irrigation conditions.
Study Design: Landrace varieties from different regions of Iran were selected for evaluating the interrelationship of traits under drought stress.
Place and Duration of Study: The Research Farm of Department of Crop Production and Plant Breeding, College of Agriculture, Shiraz University, Shiraz, Iran, between 2010 and 2012 growing seasons.
Methodology: Thirty five wheat genotypes consisting of 33 landrace varieties and two cultivars were cultivated as a split plot design in three replications in 2010-11 and 2011-12 growing seasons. Drought stress and normal irrigation conditions were considered as main plots and genotypes were cultivated in subplots.
Results: Cryptic relationships among antioxidants and agronomic traits were defined by 7 and 6 factors that explained 80% and 75% of traits variation under fully irrigated and drought stress conditions respectively. Factor 2 was defined as grain yield factor and it was a contrast between antioxidants and morphological traits. In factor 2, grain yield, thousands grain weight, spikelet and grain number had the highest loadings. Stepwise

**Corresponding author: Email: bheidari@shirazu.ac.ir;*

regression for grain yield (Y) and other traits under drought stress indicated that thousand grain weight (X_1), biological yield (X_2), harvest index (X_3) and grain number (X_4) entered to grain yield model as $Y= 44.4+ 3.03 X_1+ 0.389 X_2+ 12.635 X_3+ 2.639 X_4$. Except day to heading and canopy temperature, agronomic traits had positive correlations with grain yield. Cluster analysis showed that genotypes assigned to 5 clusters under drought stress and the highest grain yield (5.3 t ha^{-1}) and harvest index (38.1%) belonged to the fifth cluster. The genetic coefficients of variation were from 1.3% (superoxide dismutase) to 19.0% (thousand grain weight) in 2010-11 and 1.52% (superoxide dismutase) to 17.2% (protein) in 2011-12. Canonical analysis showed that canopy temperature and heading were negatively associated with grain yield and with superoxide dismutase (SOD) and catalase (CAT) as antioxidant enzymes under drought stress conditions.
Conclusion: Antioxidants and canopy temperature had lowest genetic advance under drought but thousand grain weight and harvest index had highest genetic advance and can be considered as selection index for drought tolerance improvement.

Keywords: Drought; genetic advance; landrace; multivariate analysis; wheat.

1. INTRODUCTION

Growing population in developing countries accompanied with high food consumption in developed countries lead to high global demand on food consumption [1]. Massive using of bred lines or synthetic cultivars results in germplasm uniformity and consequently crop vulnerability against biotic and abiotic stresses [2,3,4,5]. In Iran, the state of Fars which is in the first rank of wheat production experienced long time drought stress due to low precipitation. Average wheat grain yield in Fars has been around 2 t ha^{-1} [6]. Expanding genetic diversity guarantees feeding world population and crop germplasm against adverse effects of environmental stresses. Variations in crop germplasms also increase the chance of finding high yielding plants for cultivation under unfavorable environmental conditions. Screening various germplasms is the first step of a basic program for selection of candidate genotypes for cultivation under targeted regions. One of the rich genetic resources are landrace varieties harboring valuable genes against adverse effects of biotic and abiotic stresses [7]. A landrace is a local variety of a domesticated plant species which has developed largely by natural processes, by adaptation to the natural and cultural environment. Landrace differs from a formal breed which has been selectively bred deliberately to conform to a particular trait. Landraces are usually more genetically and physically diverse than formal breeds. The features of landrace varieties could be incorporated into commercial cultivars and inbred lines by under field hybridizations or *In vitro* transformation methods.

Evaluation of the interrelationship of traits in crop germplasm is important for clarifying population structure and modeling selection criteria for increasing plant productions. Simple statistical analyses are not able to clarify cause and effect relations of important traits contributed to grain yield variations. Alternatively, multivariate analyses provide a comprehensive view of the interrelationship between traits that can efficiently be used in modeling population structure and crop production [8,9]. Simple correlation coefficients show the extent and direction of relation of a pair trait although indirect effects of other traits cannot be determined. Alternatively, by path analysis the simple correlation coefficients could be partitioned into direct and indirect effects of a trait on grain yield [10]. Path analysis is a special case of structural equation or causal modeling, but no measurement modeling. Stepwise linear regressions proved to be more efficient than simple regression models to

determine the predictive equation of grain yield [11,12]. Stepwise regression allows selecting the most prominent traits that significantly affect grain yield by excluding non significant ones. Traits entered to the model of stepwise regression can be used for path coefficients analysis and scoring direct and indirect effects on grain yield. Deliberately selection of parental lines in hybridization programs is the first step in plant breeding. In order to benefit from hybrid vigour or prominent segregates, information of similarities between parents is necessary [13]. Clustering is a multivariate analysis that uses a combination of traits or genotypes information to classify a population into main groups based on similarities [14,15]. Factor analysis is a statistical method used to describe variability among correlated variables in terms of a potentially lower number of unobserved variables called factors. Factor analysis searches for joint variations in response to unobserved latent variables. The observed variables are modelled as linear combinations of the potential factors [11,12,16]. Further information about the interdependencies between observed variables can be used later to reduce the set of variables in a dataset. Selecting genotypes based on loading factors in factor analysis increases response to selection and makes breeders to be sure about considering important traits for increasing grain yield.

Information of genetic parameters is also important in modeling selection approaches for improving crop structures [17,18,19]. Estimating heritability of traits and the extent of genetic variation can be used in prediction of genetic advances in a crop population [19]. Therefore, given the importance of genetic variation and resolution of population structure for crop improvement, the main objectives of this study were to investigate the interrelationships of agronomic and biochemical traits and the extent of genetic variation in wheat landrace varieties versus commercial cultivars by using multivariate analyses under fully irrigated and drought stress conditions. Genotypes were evaluated in two years in order to determine changes in traits and the effects of year on relationships between traits.

2. MATERIALS AND METHODS

2.1 Plant Materials and Field Condition

The experiment was conducted during 2010-11 and 2011-12 growing seasons. The site of experiment located at the Research Farm of Shiraz University, Shiraz, Iran. Thirty five wheat cultigens consisted of 2 commercial cultivars (Shiraz and Cross-bulani) and 33 landrace varieties were selected for screening drought tolerance. Shiraz is a commercial cultivar adapted to water limited conditions and is cultivated in some of regions in Fars, Iran. Cross Bulani is also used as drought tolerance control. Selected varieties were KC4565 (1), KC4568 (2), KC4818 (3), KC4500(4), KC4548 (5), KC4864 (6), KC4617 (7), KC2194 (8), KC3892 (9), KC4847 (10), KC4567 (11), KC2172 (12), KC4557 (13), KC4495(14), KC3893 (15), KC4633 (16), KC4604 (17), KC2177 (18), Cross Bulani (19), KC4619 (20), KC4618 (21), KC4537 (22), KC4542 (23), KC4862 (24) KC4543 (25), KC3885 (26), KC2165 (27), KC4929 (28), KC4595 (29), KC3878 (30), KC3891 (31), Shiraz (32), KC4512 (33), KC4492 (34), KC4551(35). The landrace cultigens collected from different regions of Iran by the National Seed and Plant Improvement Institute, Iran. Landraces are highly variable morphologically. The KC preceded in the name of landraces refers to Karaj Center, for Agricultural Research, Karaj, Iran.

Experimental design was a split plot based on randomized complete block design with 3 replications. One of main plots allocated to fully irrigated water regime and the other assigned to drought stress. Genotypes were sown in each main plot. Each experimental plot

had 3 rows 2 m long. Prior to sowing, the field was fertilized with 50 kg N ha^{-1} and 110 kg ha^{-1} triple superphosphate. On November 2010 and 2011, the seeds of the genotypes were sown at a depth of 5 cm with density of 350 seed m^{-2}. During the growing season, 50 kg N ha^{-1} was added to the soil at each of the stem elongation and heading stages. The soil texture was sandy clay with pH 7. Weeds were controlled by using the herbicide "Total" (40 g ha^{-1} sulfosulfuron (75%) + met sulfuron methyl (5%)) at the tillering stage. Hand- pulling of weeds was also used throughout the growing season. Under fully irrigated condition, genotypes were fully watered (field capacity) throughout growing season until it was necessary. Under drought stress, plants fully watered until the time that 50% of spikes of each of genotypes emerged from leaf sheaths and from that time irrigation was stopped until end season.

2.2 Data Collection and Measurements

2.2.1 Protein content

Leaf samples were taken from plants at grain filling stage for quantifying total protein [20].

2.2.2 Proline

Proline content was determined by a modification in Bates et al. [21] procedure. The leaves were homogenized in 2 ml of 3% sulfosalicylic acid solution using tissue homogenizer. The homogenate was then centrifuged at 13000 g for 10 min. Supernatant (1 ml) was then added into a test tube contained 1 ml of glacial acetic acid and 1 ml of freshly prepared acid ninhydrin solution (1.25 g ninhydrin dissolved in 30 ml of glacial acetic acid and 20 ml of 6 M orthophosphoric acid). Tubes were incubated in a water bath for 1 h at 100 ℃. Two ml of toluene was added and mixed on a vortex mixture for 20 s under hood. After 10 min, toluene separated from aqueous phase in test tubes. The absorbance of toluene phase was measured at 520 nm with spectrophotometer.

2.2.3 Ascorbate peroxidase (APX)

APX activity was measured using the method of Nakano and Asada [22]. The assay mixture was 50 mM potassium phosphate buffer (pH 7.0) containing 0.5 mM ascorbic acid, 0.15 mM H_2O_2, 0.1 mM EDTA, and 50 µL of enzyme extract (supernatant). Ascorbate peroxidase (U g^{-1} fresh weight (FW)) was spectrophotometrically assayed following a decrease in the absorbance at 290 nm. One unit (U) of APX oxidises 1 mM ascorbic acid in 1 min at 25℃.

2.2.4 Catalase (CAT)

CAT activity (U g^{-1} FW) was measured by following the reduction of H_2O_2 at 240 nm according to the method of Dhindsa et al. [23]. The starting solution contained 50 mM potassium phosphate buffer (pH 7.0) and 15 mM H_2O_2. The reaction was started by adding 100 µl enzyme extract to the reaction mixture and changes in absorbance were determined 1 min after the start of the reaction. One unit of activity CAT was considered as the amount of enzyme which decomposes 1 mM of H_2O_2 per minute.

2.2.5 Peroxidase (POD)

POD activity (U g^{-1} FW) was determined according to the method of Chance and Maehly [24]. The enzyme was assayed in a solution containing 50 mM phosphate buffer (pH 7.0), 5

mM H_2O_2 and 13 mM guaiacol. The reaction was initiated by adding 33 µl enzyme extract at 25°C. One unit of enzyme was calculated based on the formation of tetraguaiacol in 1 min. Tetraguaiacol has a maximum absorption at 470 nm and its reaction can be readily followed spectrophotometrically.

2.2.6 Superoxide dismutase (SOD)

The reaction mixture contained 50 mM phosphate buffer (pH 7.8), 0.1 mM EDTA, 13 mM methionine, 75 µM nitroblue tetrazolium (NTB), 2 µM riboflavin and 100 µl of the supernatant [25]. Riboflavin was added as the last component and the reaction was initiated by placing the tubes under two 15 W fluorescent lamps. The reaction was terminated after 15 min. Non-illuminated and illuminated reactions without supernatant served as calibration standards. Products were measured at 560 nm. One unit of SOD activity (U g^{-1} FW) was defined as the amount of enzyme that inhibited 50 nitroblue tetrazolium (NBT) photoreduction.

2.2.7 Agronomic traits

Number of day to heading was counted based on the difference of sowing date and the time that spikes emerged from 50% of plants in each plot. Plant height was measured from the ground level to the tip of a spike during the grain filling stage. After pollination, 10 plants per plot were selected for measuring number of spikelet per spike, spike length, canopy temperature and peduncle length. After harvesting plants, grain per spike and thousand grain weight were measured. Grain yield (GY) at 14% moisture content and biological yield (BY) were measured as per square meter that transferred to t ha^{-1}. Harvest index (HI) was calculated by using the following equation:

$$HI = \frac{GY}{BY} \times 100$$

2.3 Data Analysis

Heritability in broad sense (h^2) was estimated using following formula [26]:

$$h^2 = \frac{\sigma_g^2}{\sigma_p^2} \times 100$$

Where, σ^2_g and σ^2_p are respectively genotypic and phenotypic variance components. Genotypic and phenotypic variances were estimated using expected mean squares of genotype (MSg) and error (MSe) from the analysis of variance [27] as bellow:

$$\sigma_g^2 = \frac{MS_g - MS_e}{r} \qquad \sigma_p^2 = \sigma_e^2 + \sigma_g^2 \qquad \sigma_e^2 = \frac{MS_e}{r}$$

Where, r is the number of replication. Phenotypic (PCV) and genetic (GCV) coefficients of variation were calculated based on the mean ($\bar{x}$) of each trait as follow (Falconer 1989):

$$GCV = \frac{\sqrt{\sigma_g^2}}{\bar{x}} \times 100 \qquad PCV = \frac{\sqrt{\sigma_p^2}}{\bar{x}} \times 100$$

Genetic advance (GA) for each trait was estimated as bellow [26]:

$$GA = K \times \sigma_{ph} \times h^2$$

In this formula, K (= 2.06) is selection intensity at 5% and σ_{ph} is the square root of phenotypic variance. In order to compare GA of traits, genetic advance based on mean (GAM) was also calculated by dividing GA to the mean of each trait. Analysis of data and variances (ANOVA) was performed using SAS 9.2 [28] software.

To perform factor analysis, principal component analysis (PCA) was used for factor extraction. Factor weights were computed in order to extract the maximum possible variance, with successive factoring continuing until there is no further meaningful variance left. The factor loadings were then rotated for better interpretation of factors [29].

Canonical correlation analysis, developed by Hotelling [30], often allows a more meaningful interpretation of interrelations between variables than simple correlation analysis. Canonical correlation analysis utilizes 2 sets of variables and forms linear indices from each of the sets of variables so that the correlation between the two indices is maximized. Several pairs of linear indices are derived and are named canonical variables. Specifically, the ith pair of canonical variable is described as:

$$U_i = \sum^{p} a_{ij} z_j$$

$$V_i = \sum^{q} b_{ij} z_j$$

, i = 1,2,3…p and j= 1,2,3, …q

where $q \geq p$, z_j is the jth variable, p is the number of variables in the first set, q is the number of variables in the second set, a_{ij} and b_{ij} are the variable coefficients (or loadings) in the first and second sets, respectively. In this study we used agronomic (Agr) traits and antioxidants (Ant) as two sets of traits for analysis of canonical correlations and better interpreting of the interrelationships between traits under drought stress over two years. To do this, we used agronomic traits as U variable and antioxidants as V variable based on combined data of both years. Correlations of each of original agronomic traits with their canonical variable (Agr) and with cross (Ant) canonical variable were calculated. This procedure was applied for antioxidants as original traits with their canonical variable (Ant) and the other cross (Agr) canonical variable.

Cluster analysis [29] was used for grouping 35 wheat genotypes into sets of genotypes with closest distances and similarities. All traits were used for constructing similarity matrix and clustering genotypes under fully irrigated and drought stress conditions. Stepwise regression was used to define a model of traits that are highly contributed to grain yield predictions. To do stepwise regression, grain yield (Y_i) was considered as dependent that regressed on all traits (X_i) as independent at first step. Hieratically, non-significant traits that had no significant contribution to grain yield variations were excluded from the model.

3. RESULTS AND DISCUSSION

3.1 Grain yield under Drought Stress and Fully IrrigatedTrials

Grain yield varied under drought stress and fully irrigated trials as affected by different conditions of two years. Drought stress at reproductive stages decreases grain set in wheat by inducing pollen sterility [31]. Genotypes had lower grain yield under drought tress compare to fully irrigated conditions (Table 1). Mean values for grain yield were higher in 2010-11 trial as compared to the means in 2011-12 growing season. Mean grain yield ranged from 6.8 t ha^{-1} in fully irrigated trial in 2010-11 to 2.40 in 50% heading drought stress in 2011-12. The lowest mean (2.40 t ha^{-1}) was belonged to KC3878 in 50% heading drought stress trial which was against the highest (9.17 t ha^{-1}) in KC2165 in fully irrigated trial (2010-11).

Table 1. Mean grain yield (t ha^{-1}) under 4 trials in 2010-11 and 2011-12 growing seasons in wheat genotypes

Varietiy/Cultivar	FI (2010-11)	50% HDS (2010-11)	FI (2011-12)	50% HDS (2011-12)
KC4565	6.73	5.05	5.40	4.13
KC4568	7.34	5.05	6.60	4.53
KC4818	6.70	5.13	4.33	3.07
KC4500	6.46	4.46	4.60	4.11
KC4548	6.40	4.46	5.57	3.97
KC4864	7.00	5.16	5.53	3.40
KC4617	5.21	3.52	4.13	3.83
KC2194	6.77	5.02	5.30	2.70
KC3892	6.61	5.97	4.83	3.13
KC4847	4.70	3.60	3.87	2.67
KC4567	6.99	4.77	4.67	3.60
KC2172	7.24	4.80	5.83	4.10
KC4557	8.64	7.27	7.23	4.97
KC4495	7.81	6.34	5.03	4.57
KC3893	5.81	4.68	6.10	4.63
KC4633	8.14	7.14	6.93	5.40
KC4604	5.17	4.94	4.10	3.33
KC2177	7.31	5.70	6.37	4.20
Cross Bulani	6.44	5.86	6.00	4.23
KC4619	6.84	6.22	5.70	4.13
KC4618	6.43	5.39	5.13	4.40
KC4537	7.90	7.17	6.43	5.12
KC4542	7.39	5.19	7.57	5.83
Kc4862	8.16	7.47	7.07	5.33
KC4543	7.21	4.81	6.63	3.77
KC3885	6.29	5.35	6.53	4.47
KC2165	9.17	5.41	6.10	4.50
KC4929	5.57	4.80	6.13	4.30
KC4595	5.54	3.54	5.77	3.07
KC3878	4.60	3.87	3.07	2.40
KC3891	8.46	7.93	7.03	5.50
Shiraz	5.81	3.82	4.33	3.93
KC4512	6.86	5.13	6.03	4.57
KC4492	6.80	5.26	6.00	4.13
KC4551	7.46	6.34	6.76	5.33
Mean	6.8	5.3	5.7	4.1

HDS: 50% heading drought stress, FI: fully-irrigated

3.2 Factor Analysis Results for Cryptic Relationship between Traits

Results of factor analysis for fully irrigated and drought stress conditions are presented in Table 2 and 3 respectively. Changes in eigen values plotted against number of factors (Fig. 1). Fig. 1 shows that eigen values were not significantly changed after factor 7. Therefore, 7 factors were selected for the interpretation of cryptic relationships between traits under fully irrigated conditions. Seven selected factors cumulatively accounted for 80% of the total variations of traits under fully irrigated condition of both years. The first factor mainly defined by grain yield, grain number per spike, thousand grain weight, harvest index and number of spikelet per spike under fully irrigated conditions. This factor accounted for 24% of the total variation and could be defined as grain yield factor. Therefore, selection of genotypes based on loadings in this factor increase grain yield and its components. Loading factors of fertile tiller and biological yield were higher than loadings of other traits in factor 2 therefore, this factor can be named as biological yield and selections based on this factor increases the size of crop and total dry matter. The third factor clearly separated two groups of biochemical traits. This factor which includes total protein (loading= 0.85) and proline (loading= 0.85) accounted for 12% of the total variation of all traits. The fourth and fifth factors could be named as spike feature and ascorbate peroxidase respectively. Factor 6 was strongly influenced by number of fertile tiller and canopy temperature. Therefore, this factor increases tillers and canopy temperature of the wheat cultivars under fully irrigated conditions. Loading coefficients in factor 7 indicated that canopy temperature with loading equal to 0.51 has significant contribution to total variance of this factor. Selections based on loadings of this factor increase canopy temperature.

Table 2. Factor analysis based on antioxidants and agronomic traits in wheat genotypes in fully irrigated condition

	Eigen vectors						
Traits	**Factor1**	**Factor2**	**Factor3**	**Factor4**	**Factor5**	**Factor6**	**Factor7**
Plant height	0.37	-0.42	0.06	-0.12	0.01	0.18	-0.55
Canopy temperature	-0.37	-0.22	-0.14	-0.40	-0.07	0.44	0.51
Spike length	0.00	0.04	-0.19	0.63	0.15	0.38	-0.25
Peduncle length	0.30	0.33	0.40	-0.44	0.32	-0.38	-0.12
Fertile tiller	-0.10	0.52	0.44	-0.19	-0.04	0.67	-0.06
Number of spikelet per spike	0.86	-0.23	0.02	0.04	0.03	0.21	0.08
Grain number per spike	0.80	0.09	-0.26	-0.19	-0.11	0.07	0.21
Thousand grain weight	0.94	-0.16	-0.14	-0.10	-0.03	0.11	-0.01
Biological yield	0.45	0.66	0.09	-0.31	-0.12	0.10	-0.28
Harvest index	0.65	-0.54	0.05	0.34	0.05	-0.13	0.18
Day to heading	-0.15	-0.51	-0.16	-0.25	-0.23	0.20	-0.16
Grain yield	0.96	0.30	0.01	0.06	-0.06	-0.02	-0.01
Protein	0.06	-0.28	0.85	0.13	-0.05	0.01	0.01
Proline	0.08	-0.20	0.85	0.12	-0.18	0.10	0.20
Peroxidase	0.23	0.48	-0.06	0.33	-0.55	-0.11	0.28
Ascorbate peroxidase	0.35	0.29	-0.52	-0.13	0.41	0.09	0.27
Catalase	-0.01	0.41	0.15	0.35	-0.29	0.13	-0.12
Superoxide dismutase	0.02	0.13	0.19	0.31	0.79	0.19	0.12
Eigen value	4.39	2.63	2.25	1.51	1.42	1.18	1.04
Proportion of variance explained	0.24	0.15	0.12	0.08	0.08	0.07	0.06
Cumulative percentage of variance	0.24	0.39	0.52	0.60	0.68	0.74	0.80

Under drought stress conditions, eigen values plotted against number of factors (Fig. 2). Fig. 2 shows that from the factor sixth there is a slight change in eigen values and the first 6 factors that totally explained 75% of total variation could be selected for the interpretation of interrelationships of traits Table 3. The first factor had the highest eigen value (5.38) and explained 30% of total variations under drought stress. Grain yield, thousands grain weight, spikelet per spike and grain number per

Table 3. Factor analysis based on antioxidants and agronomic traits in wheat genotypes in drought stress condition

	Eigen vectors					
Traits	**Factor 1**	**Factor 2**	**Factor 3**	**Factor 4**	**Factor 5**	**Factor 6**
Plant height	0.35	0.06	-0.69	0.03	-0.08	-0.14
Canopy temperature	-0.34	0.41	0.36	-0.09	0.03	0.35
Spike length	0.33	-0.15	-0.22	0.29	0.62	-0.06
Peduncle length	0.50	-0.29	0.05	-0.52	-0.17	-0.34
Fertile tiller	0.25	-0.37	-0.07	-0.45	-0.12	0.51
Number of spikelet per spike	0.78	0.39	-0.15	-0.05	0.12	0.12
Grain number per spike	0.70	0.43	-0.23	0.06	0.29	0.00
Thousand grain weight	0.85	0.41	-0.08	0.06	0.00	0.05
Biological yield	0.68	0.05	0.20	-0.54	0.16	0.17
Harvest index	0.61	0.32	-0.09	0.32	-0.39	-0.18
Day to heading	-0.24	0.43	-0.41	0.18	0.08	0.27
Grain yield	0.87	0.33	0.04	-0.10	-0.14	-0.04
Protein	0.44	-0.63	-0.17	0.25	-0.21	0.20
Proline	0.37	-0.58	-0.18	0.33	-0.20	0.26
Peroxidase	0.36	0.03	0.47	0.29	-0.06	0.44
Ascorbate peroxidase	0.48	-0.04	0.45	0.26	0.19	-0.31
Catalase	0.46	-0.37	0.45	0.12	-0.37	-0.16
Superoxide dismutase	0.47	-0.51	0.09	-0.04	0.49	-0.01
Eigen value	5.38	2.43	1.96	1.35	1.24	1.12
Proportion of variance explained	0.30	0.13	0.11	0.08	0.07	0.06
Cumulative percentage of variance	0.30	0.43	0.54	0.62	0.69	0.75

spike had the highest loading coefficients in the first factor. Therefore, factor 1 could be defined as grain yield component. Factor 2 was a contrast between antioxidant enzymes and morphological traits. As a consequence, selection of genotypes using loading coefficients of factor 2 reduces biochemical traits under drought stress and slightly changes some of morphological traits. Factor 3 was a contrast between biochemical traits, plant height and heading date. Coefficients of traits in third factor show that antioxidant enzymes increased under drought stress but plant height and number of day to heading reduced. In factor 4, peduncle length, fertile tiller and biological yield had negative coefficients which were against biochemical traits with positive loadings. The fifth factor was strongly affected by spike length coefficient and could be named as spike feature factor. Fertile tiller, canopy temperature and peroxidase had positive and higher coefficients than other traits in factor 6. Therefore, it could be concluded that increase in the number of fertile tillers increase canopy temperature under drought stress. Mohamed [12] used factor analysis to classify ten wheat variables into

two main groups which accounted for 80.79% of the total variability in the dependence structure.

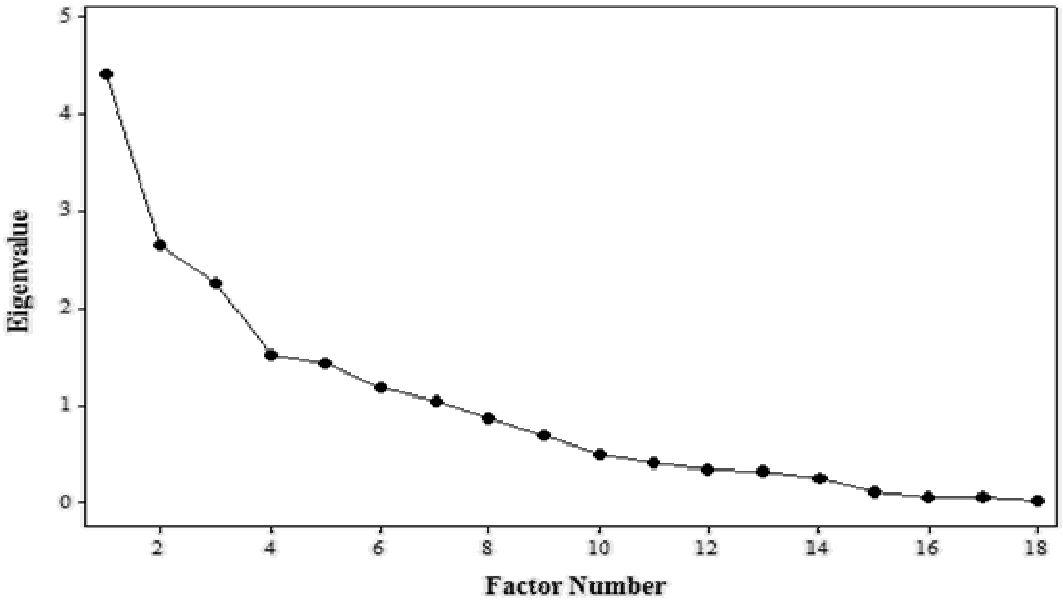

Fig. 1. Projection of the number of factors against their eigen values under fully irrigated condition

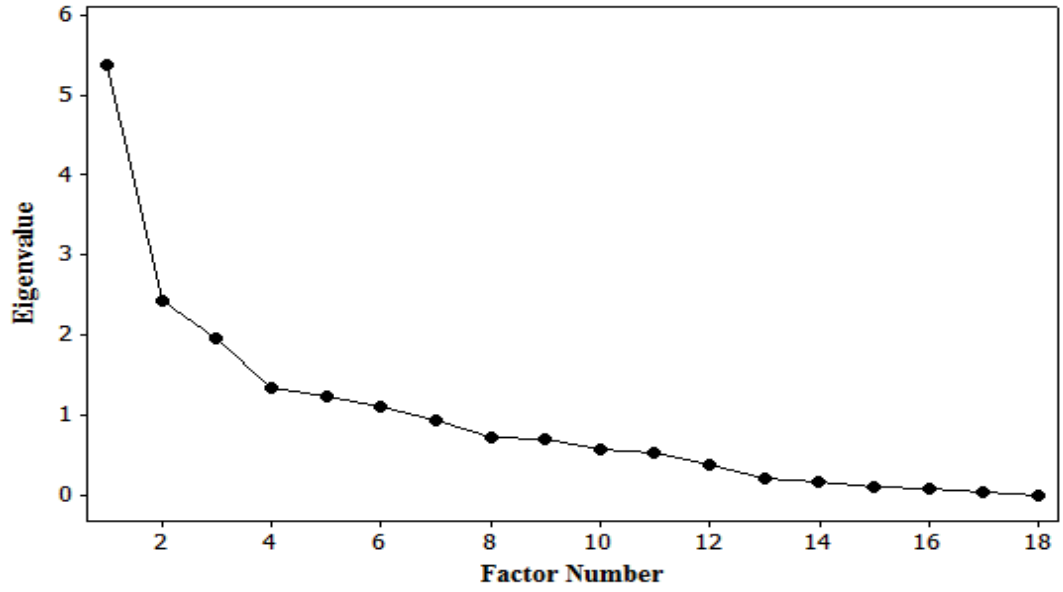

Fig. 2. Projection of the number of factors against their eigen values under drought stress condition

3.3 Modeling Grain yield under Fully Irrigated and Drought Conditions

Results of stepwise analysis are shown in Table 4. In fully irrigated conditions, thousand grain weight (R^2= 79.8%), biological yield (R^2= 7.8%), harvest index (R^2= 1.3%) and spike length (R^2= 0.4%) entered to the model as significant traits that highly contributed to grain yield variations. These traits explained 89.3% of total variations of grain yield under fully irrigated conditions. The model of stepwise regression under fully irrigated condition was as below:

$$Y= 44.4+ 5.4\ X_1+ 11.391\ X_2+0.307\ X_3+5.767\ X_4$$

In this model Y, X_1, X_2, X_3 and X_4 denote for grain yield, thousand grain weight, harvest index, biological yield and spike length respectively.

Table 4. Stepwise regression models for grain yield (Y) prediction in wheat genotypes under fully irrigated and drought stress conditions

Variable Entered	Fully irrigated trial				
	Parameter Estimate	Partial R-Square	Model R-Square	F Value	Pr > F
Thousand grain weight (X_1)	5.4002	0.798	0.798	130.85	<.0001
Harvest index (X_2)	11.391	0.013	0.811	3.53	0.0402
Biological yield (X_3)	0.3074	0.078	0.889	56.56	<.0001
Spike length (X_4)	5.7673	0.004	0.893	3.15	0.0362

Grain yield prediction in fully irrigated trial: $Y= 44.4+ 5.4002\ X_1+ 11.391\ X_2+0.307\ X_3+5.7673\ X_4$

Variable Entered	Drought stress trial				
	Parameter Estimate	Partial R-Square	Model R-Square	F Value	Pr > F
Thousand grain weight (X_1)	3.02	0.697	0.697	76.2	<.0001
Biological yield (X_2)	0.3893	0.099	0.796	14.35	0.0006
Harvest index (X_3)	12.635	0.098	0.894	628.04	<.0001
Grain number (X_4)	2.639	0.008	0.902	3.16	0.0052

Grain yield prediction in drought stress trial: $Y= 44.4+ 3.03\ X_1+ 0.3893\ X_2+ 12.635\ X_3+ 2.639\ X_4$

The model of stepwise regression for grain yield and other traits under drought stress indicated that thousand grain weight, biological yield, harvest index and grain number per spike had higher contributions to grain yield variations. Drought at early stage of heading and during grain filling decreases the number of endosperm cells and number of starch granules per cell which consequently decrease grain size and weight in wheat genotypes [31]. The model (R^2= 90.2%) for grain yield (Y) and entered traits including thousand grain weight (X_1), biological yield (X_2), harvest index (X_3) and grain number per spike (X_4) was as follow:

$$Y= 44.4+ 3.03\ X_1+ 0.389\ X_2+ 12.635\ X_3+ 2.639\ X_4$$

This model shows that regression coefficients are positive and all entered traits had significant effects on grain yield.

3.4 Association of Traits

Simple correlation coefficients of agronomic traits are presented in Table 5. The intensity of correlation coefficients was lower under drought compared to irrigated conditions showing the effect of drought on the interrelationship of traits. Except number of day to heading and

canopy temperature, other agronomic traits had positive correlations with grain yield in both years. Grain yield had higher correlations with thousand grain weight, harvest index, spikelet per spike under both fully irrigated and drought stress conditions. Canopy temperature had significantly negative correlations with grain yield under drought stress in both growing seasons. The weight of harvested total grain as a percentage of total plant weight of the crop is an important character that can be used for selection of high yielding plants. In most cases, the improvement in harvest index has been a consequence of increased grain population density coupled with stable individual grain weight [32]. In present study, harvest index was significantly correlated with thousand grain weight and grain number per spike under drought stress. In both years, biological yield was significantly correlated with spike length and number of fertile tiller. Positive correlations of fertile tiller with grain yield and grain per spike with spikelet per spike have been reported by Mondal et al. [33] and Raut et al. [34].

The first two factors obtained from factor analysis were plotted to assess the interrelationships of traits under drought stress conditions Fig. 3. Angles between traits vectors showed that thousand grain weight, grain number per spike, spikelet per spike and harvest index were highly associated with grain yield. Therefore, these traits are adapted to drought conditions. The angles between canopy temperature and day to heading vectors with grain yield indicated that they were not associated with grain yield. Wide angles between antioxidants and grain yield vectors show that antioxidants are not correlated with grain yield under drought stress conditions.

3.5 Canonical Analysis

The canonical variable loadings and the correlations of the original variables with the canonical variates are valuable in interpreting the interrelations between the canonical variates. Results of canonical analysis for drought trials are presented in Tables 6. Results showed that the correlation of the first pair of the canonical variables was relatively high (0.98**) but the second pair had lower correlation (0.74). Therefore, interpreting the first pair canonical variable is more focused. Estimated canonical loadings showed that the agronomic (Agr) canonical variable had the largest absolute coefficients on grain number, fertile tiller and grain yield while in the first antioxidant (Ant) canonical variable, SOD and CAT had the highest loading coefficients. Grain yield (r= 0.51) and spike length (0.56) had higher correlation with Agri1 canonical variable while heading and canopy temperature were negatively associated with Agri1. This shows that higher canopy temperature at grain filling reduces end use grain yield under drought stress condition. The first Ant canonical variable had higher correlation with SOD, proline and CAT but it had negative correlation with protein content under drought stress condition. This result shows that wheat genotypes with higher grain yield accumulated higher antioxidants but their protein content was reduced as a result of drought. Therefore, correlation coefficients in the first pair of canonical variables show that different antioxidants have unequal contributions to drought stress tolerance and that canopy temperature reduced antioxidant accumulations under drought stress conditions. Cross-correlations of Ant canonical correlation with agronomic traits indicated that Ant1 variable was negatively correlated with heading and canopy temperature while Agri1 had negative correlation with protein content. This shows that when wheat genotypes spend their energy for increasing agronomic drought- adaptive traits they will not have enough energy for accumulating protein under drought conditions.

Table 5. The correlation coefficients of agronomic traits under drought stress (in parenthesis) and fully irrigated conditions in 2010-11 (under diagonal) and 2011-12 (above diagonal)

Traits	GY	BY	HI	TGW	GN	SPS	PH	NDH	CT	SL	PL	FT
GY	1	0.61**	0.70**	0.95**	0.65**	0.63**	0.32	-0.13	-0.59*	0.57**	0.44*	0.56**
		(0.59**)	(0.61**)	(0.88**)	(0.60*)	(0.66**)	(0.02)	(-0.18)	(-0.48*)	(0.67**)	(0.32)	(0.45*)
BY	0.22	1	-0.34	-0.16	0.35	0.41*	0.13	0.34	0.12	0.55*	0.54*	0.56*
	(0.32)		(-0.30)	(0.15)	(0.27)	(0.47*)	(0.45*)	(0.22)	(0.11)	(0.44*)	(0.43)	(0.43*)
HI	0.63**	0.34*	1	0.56*	0.45**	0.34	0.34	-0.34	-0.45*	0.34	0.12	0.43
	(0.71**)	(-0.16)		(0.44**)	(0.39*)	(0.44*)	(0.09)	(0.01)	(0.33)	(0.38)	(0.11)	(0.23)
TGW	0.88**	0.33	0.54*	1	0.81**	0.84**	0.28	-0.03	0.23	0.33	0.18	0.33
	(0.72**)	(0.42*)	(0.48*)		(0.63**)	(0.55**)	(0.08)	(-0.13)	(-0.13)	(0.28)	(0.12)	(0.34)
GN	0.59**	-0.02	0.45*	0.76**	1	0.64**	0.16	0.04	0.19	0.78**	0.15	0.55*
	(0.36*)	(0.24)	(0.43*)	(0.88**)		(0.61**)	(0.12)	(0.01)	(0.09)	(0.59**)	(0.13)	(0.43*)
SPS	0.67**	0.67**	0.46*	0.82**	0.78**	1	0.20	0.01	-0.24	0.68*	0.33	0.45
	(0.59**)	(0.43*)	(0.14)	(0.59*)	(0.69**)		(0.06)	(-0.07)	(-0.04)	(0.47*)	(0.23)	(0.36)
PH	0.26	0.11	0.11	0.19	0.03	0.10	1	0.13	0.12	0.32	0.34	0.23
	(0.12)	(0.18)	(0.06)	(0.21)	(0.12)	(0.32)		(0.10)	(0.11)	(0.18)	(0.41)	(0.28)
NDH	-0.04	0.23	-0.07	-0.06	0.01	0.15	0.21	1	-0.09	0.23	0.13	0.23
	(-0.21)	(0.25)	(0.04)	(-0.02)	(0.09)	(0.03)	(0.09)		(-0.15)	(0.22)	(0.09)	(0.21)
CT	-0.76*	0.13	0.08	0.21	-0.24	-0.16	0.01	-0.32	1	0.09	-0.11	-0.45**
	(-0.43*)	(0.23)	(0.04)	(-0.04)	(0.02)	(-0.12)	(0.12)	(-0.09)		(0.14)	(0.03)	(-0.34)
SL	0.45*	0.49*	0.27	0.30	0.46*	0.66**	0.19	-0.02	0.21	1	0.34	0.31
	(47*)	(0.42*)	(0.32)	(0.27)	(0.38*)	(0.49*)	(0.12)	(0.10)	(0.18)		(0.27)	(0.22)
PL	0.57*	0.46*	0.16	0.14	0.38	0.33	0.12	-0.2	-0.28	0.34	1	0.21
	(44*)	(0.33)	(0.12)	(0.12)	(43*)	(0.31)	(0.09)	(0.01)	(0.03)	(0.24)		(0.18)
FT	0.56**	0.47*	0.46	0.34	0.55**	0.52**	0.26	0.25	0.09	0.30	0.33	1
	(48**)	(53*)	(0.41)	(0.27)	(0.46*)	(0.54**)	(0.13)	(0.13)	(0.11)	(0.23)	(0.21)	

** and **: significant at 5 and 1% probability level, GY: Grain yield, BY: Biological yield, HI: Harvest index, TGW: Thousand grain weight, GN: Grain number per spike, PH: plant height, SPS: Spikelet per spike, NDH: Number of day to heading, CT: Canopy temperature, SL: Spike length, PL: Peduncle length, FT: Fertile tiller*

Table 6. Standardized canonical variables (Agr and Ant) and their correlations with original agronomic and antioxidants traits in wheat genotypes under 50% heading drought stress

Variable	Coefficients in canonical variables		Correlation with their variable		Correlation with opposite variable	
Agronomy variables (Agr)	**Agr1**	**Agr2**	**Agr1**	**Agr2**	**Ant1**	**Ant2**
HD	-0.486	-0.0202	-0.53	-0.06	-0.41	-0.05
CT	-0.201	0.6798	-0.56	0.37	-0.43	0.28
PH	0.1413	-0.1044	0.26	-0.02	0.20	-0.02
FT	0.4642	0.1354	0.44	-0.08	0.34	-0.06
PL	-0.108	0.3804	-0.19	0.40	-0.15	0.29
SPL	0.2135	0.309	0.56	0.36	0.43	0.27
SPS	-0.2191	0.4846	0.41	0.08	0.32	0.06
GN	0.582	0.4444	0.46	0.00	0.35	0.00
BY	-0.298	-0.8856	0.32	-0.10	0.24	-0.08
GY	0.4249	1.0303	0.51	-0.03	0.40	-0.02
TGW	-0.0212	-0.0821	0.33	0.02	0.25	0.01
HI	-0.1514	-1.2512	0.31	-0.32	0.24	-0.23
Correlation of canonical variables	0.77	0.74				
Antioxidants variables (Ant)						
Proline	0.2075	-0.0402	0.68	0.08	0.52	0.06
Protein	-0.1359	0.9072	-0.04	0.74	-0.03	0.55
SOD	0.6829	0.3116	0.78	0.23	0.60	0.17
POD	-0.025	-0.6378	0.19	-0.34	0.15	-0.25
CAT	0.571	-0.0488	0.65	-0.23	0.50	-0.17
APX	-0.1816	-0.094	0.25	-0.27	0.20	-0.20

HD: day to heading, CT: canopy temperature, PH: plant height, FT: fertile tiller, PL: peduncle length, SPL: spike length, SPS: spikelet per spike, GN: grain number, BY: biological yield, GY: grain yield, TGW: thousand grain weight, HI: harvest index, SOD: superoxide dismutase, POD: peroxidase, CAT: catalase, APX: ascorbate peroxidase

Agri2 had no strong correlations with their original variables. Loadings in the second pair of canonical variables (Agri2 and Anti2) show that protein was increased when canopy temperature increased as a consequence of drought stress. Ant2 and Agri2 canonical variables had negative correlations with POD, CAT and APX.

3.6 Clusters of Genotypes under Fully Irrigated and Drought Conditions

Hierarchical cluster analysis was performed using the mean value of traits in fully irrigated and drought stress conditions. Genetic divergence among wheat genotypes through cluster analysis was reported by Singh and Dwivedi [35]. In this study, thirty five genotypes were grouped into five main clusters based on highest similarities under drought and fully irrigated conditions (Fig. 4 and 5).

Under fully irrigated condition, the landraces KC4565, KC4818, KC4548, KC4633, KC4618, KC3885 and KC4512 were grouped in the cluster 1 (Fig. 4). Cluster 1 had highest plant height (100.2 cm) (Table 7). Varieties of this cluster were late matured (182 days). Cluster 2 comprised of KC4568, KC4557, KC2177, KC4619, KC4537, KC4542, KC4862, KC3891 and

KC4551. This cluster had highest spikelet per spike (15.8), grain number per spike (32.7), thousands grain weight (36.4 g), harvest index (46.2%) and grain yield (7.3 t ha^{-1}). KC4604 and KC2177 assigned to cluster 3. Cluster 3 had lowest plant height, spikelet per spike, grain number per spike, thousand grain weight, harvest index and grain yield. Genotypes in cluster 3 were relatively early matured as compared to other clusters. The landrace varieties KC4500, KC3892, KC2172, KC4495, KC3893, KC4543, KC4492 and the commercial cultivar Cross Bulani joined to the fourth cluster. The fourth cluster had highest spike length. Eight genotypes consisting of KC4864, KC4617, KC2194, KC4847, KC4567, KC2165, KC4595, KC3878 and Shiraz were assigned to the fifth cluster. This cluster had lowest spike length, peduncle length, fertile tiller, biological yield and grain yield.

Under drought stress condition, cluster 1 consisted of KC4565, KC4567, KC4847, KC4595, KC4818, KC4548, KC3885 and Shiraz (Fig. 5). This cluster had lowest mean for plant height (Table 7). Cross Bulani, KC4568, KC4500, KC3892, KC4543, KC2172 grouped in cluster 2. Genotypes in this cluster had the highest plant height that significantly differed from other clusters. Five genotypes of cluster 3 consisted of KC4864, KC4617, KC2194, KC2165 and KC3878. Cluster 3 had the lowest biological (10.7 t ha^{-1}) and grain yield (3.3 t ha^{-1}). Cluster 4 constructed from KC4557, KC4495, KC4862, KC3893, KC4604, KC4551 and KC4929 and had highest biological yield (14.4 t ha^{-1}), peduncle length (20.3 cm) and fertile tiller (8.2). Cluster 4 had lowest day to heading (177.8 days) and genotypes of this group were early matured. KC4864, KC3891, KC4537, KC2177, KC4618, KC4862, KC4619, KC4542 and KC4512 joined to the fifth cluster. At this cluster, genotypes had the highest grain yield (5.3 t ha^{-1}) and harvest index (38.1%).

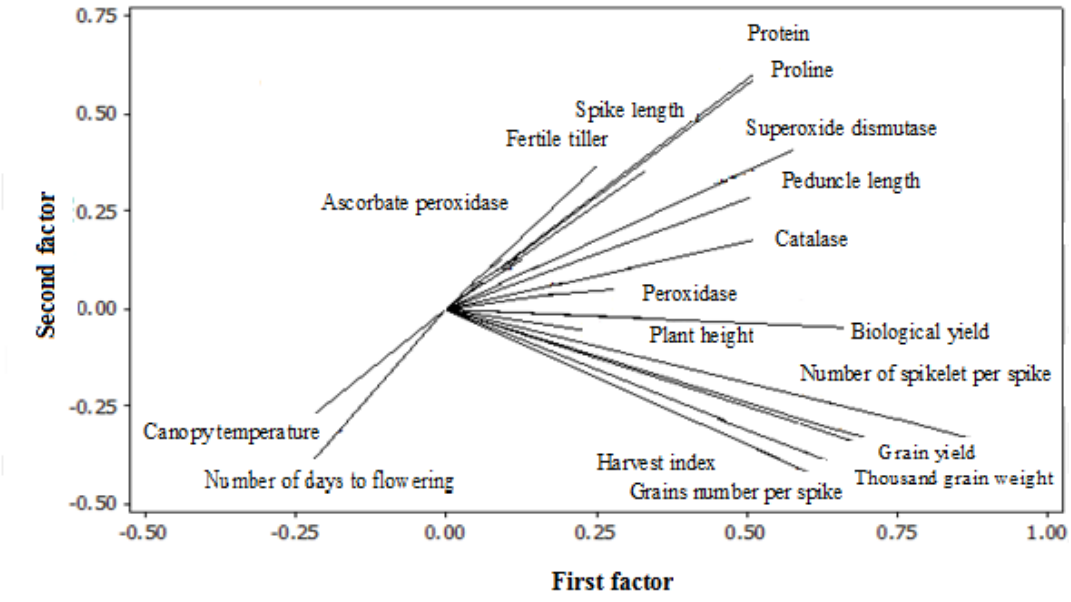

Fig. 3. Projection of traits vectors for the first and second factors that shows the association of traits under drought stress condition

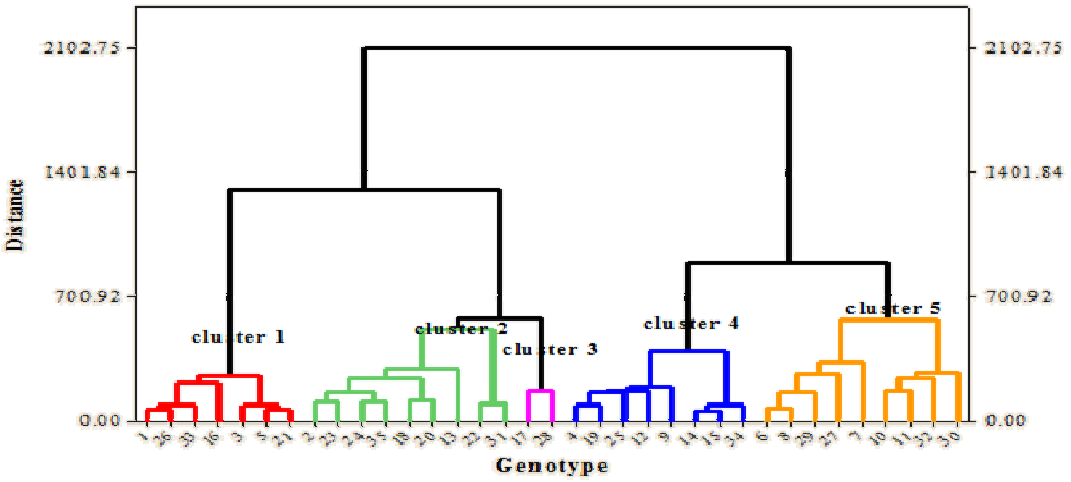

Fig. 4. Cluster diagram of 35 wheat genotypes based on agronomic traits in fully irrigated condition. Numbers refer to the name of wheat genotypes in materials and methods

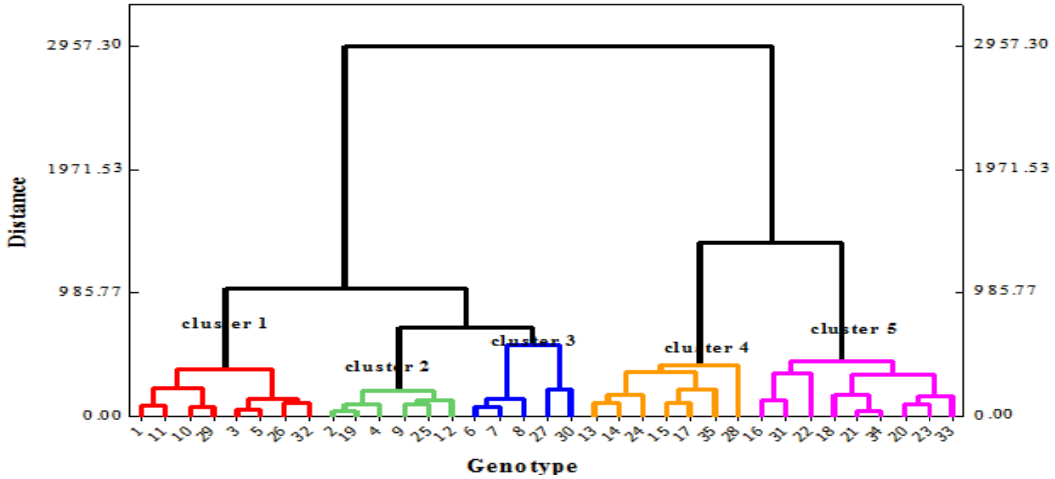

Fig. 5. Cluster diagram of 35 wheat genotypes based on agronomic traits in drought stress condition. Numbers refer to the name of wheat genotypes in materials and methods

Table 7. Mean of clusters for traits in 35 wheat genotypes under fully irrigated and drought stress (in parenthesis) conditions

Trait	Cluster					
	1	2	3	4	5	Mean square of clusters
Plant height (cm)	100.2 (96.4)	99.9 (99.2)	94.3 (96.5)	99.8 (96.6)	98.3 (96.8)	17.93** (8.84**)
Spike length (cm)	11.3 (9.5)	11.3 (10.0)	11.1 (9.9)	11.7 (10.5)	10.6(9.7)	0.29* (1.10*)
Peduncle length (cm)	20.2 (16.0)	22.24 (16.3)	23.5 (15.3)	19.7 (20.3)	19.5 (17.8)	14.60* (25.60**)
Fertile tiller	7.9 (7.1)	7.83 (7.3)	8.9 (6.7)	8.2 (8.2)	7.1 (7.3)	1.90** (1.90*)
Spikelet per spike	15.0 (13.2)	15.8 (13.6)	14.5 (13.7)	15.2 (14.36)	14.9(14.35)	1.34** (1.74**)
Grain number per spike	30.5 (25.6)	32.70 (26.0)	29.0 (26.2)	29.7 (28.3)	30.0 (28.0)	13.48** (11.15**)
Thousand grain weight (g)	32.2 (21.4)	36.42 (23.6)	25.7 (22.9)	31.5 (27.3)	29.6 (27.7)	76.74** (59.85**)
Biological yield (t ha^{-1})	14.3 (11.7)	15.5(12.2)	15.9 (10.7)	13.5 (14.4)	11.5 (14.2)	20.6** (17.2 **)
Harvest index (%)	43.3 (34.5)	46.25 (37.3)	32.8 (36.8)	45.5 (36.7)	45.6 (38.1)	81.61* (14.57**)
Day to heading (d)	182.3 (181.4)	181.1 (181.9)	178.0 (182.8)	181.0(177.8)	183.0(181.7)	13.14** (25.09*)
Grain yield (t ha^{-1})	6.2(4.0)	7.3(4.5)	5.2(3.3)	6.2(5.3)	5.4(5.4)	4.2** (3.5**)

** and ** indicate that differences of cluster are significant at 0.05 and 0.01 respectively*

3.7 Variance Components and Predictors for Genetic Advance

Genetic variance components for drought stress conditions are presented in Table 8. The PCV values were higher than GCVs. The GCV range was from 1.3% (superoxide dismutase) to 19.0% (thousand grain weight) in 2010-11 and from 1.52% (superoxide dismutase) to 17.2% (protein) in 2011-12. Lowest GCVs belonged to antioxidants and canopy temperature. This is because antioxidants are anti-stress defence metabolites synthesized *de novo* in response to drought stress and therefore antioxidants show low variations among genotypes dealt with drought conditions. Thousand grain weight, harvest index and protein in 2010-11 and protein, harvest index, fertile tiller and thousand grain weight in 2011-12 had highest GCVs respectively. This shows that morphological and agronomic traits had wider variations compared to antioxidants and canopy temperature under drought conditions.

In 2010-11, the highest heritability estimates belonged to day to heading (h^2= 88.7%) and plant height (h^2= 82.6%). In both growing seasons, antioxidants except ascorbate peroxidase had lowest heritabilities. Day to heading, spike length and ascorbate peroxidase had higher heritability in 2011-12 trial. Heritability of grain yield was 53.4% and 47.8% in 2010-11 and 2011-12 respectively. High heritability estimates for spikelet per spike, grain number, plant height, and thousand grain weight were also reported by Riaz et al. [36]. High heritability estimates indicate that selection will be effective because traits are less affected by environmental effects [37].

Heritability estimates along with genetic advance (GA) are normally more helpful in predicting genetic gain than heritability estimates alone [38]. As heritability is scale-dependent estimate, GAM was calculated by dividing GA values to the mean of each trait in a way that all GAM are comparable. Thousand grain weight and harvest index in 2010-11 and harvest index, fertile tiller and thousand grain weight had highest genetic advance in 2011-12 trial respectively (Table 8). Antioxidants and canopy temperature had lowest GAM and therefore are not recommended as index selection for increasing drought tolerance. Among grain yield components, thousand grain weight had highest GAM and it can be considered as selection index for drought tolerance improvement. Similar findings have been reported by Sharma and Garg [39]) and Dwivedi et al. [40].

Table 8. Means, lowest and highest, genotypic (GCV) and phenotypic (PCV) coefficients of variation, broad sense heritability (h^2) and genetic advance based on mean (GAM) of traits in 35 wheat genotypes under drought condition in 2010-11 and 2011-12 (in parentheses) growing seasons

Trait	Mean± SE (2010-11)	Mean± SE (2011-12)	Lowest	Highest	GCV (%)	PCV (%)	h^2 (%)	GAM (%)
Plant height (cm)	94.25±0.61	96.9±0.24	49.20 (53.9)	107.80 (103)	5.73 (3.44)	6.30 (4.98)	82.61 (47.85)	10.72 (4.91)
Canopy temperature (℃)	31.48±0.27	30.40±0.33	27.00 (28.30)	33.80 (32.30)	2.59 (2.08)	4.48 (3.07)	33.66 (45.98)	3.10 (2.91)
Spike length (cm)	9.98±0.21	9.80±0.16	5.90 (7.00)	11.80 (11.30)	8.79 (7.90)	11.51 (9.52)	58.33 (68.97)	13.83 (13.52)
Peduncle length (cm)	15.47±0.62	19.70±0.16	9.20 (12.30)	23.00 (25.40)	11.92 (3.93)	21.26 (9.47)	31.39 (52.01)	13.46 (10.14)
Fertile tiller	6.57±0.23	8.30±0.36	4.10 (5.00)	9.30 (11.33)	13.35 (13.57)	16.88 (18.55)	62.60 (53.59)	21.76 (20.47)
Spikelet per spike	13.65±0.11	15.45±0.90	13.00 (13)	15.00 (16.6)	4.78 (4.80)	7.18 (6.04)	43.75 (63.22)	6.46 (7.86)
Grains number per spike	29.05±0.27	27.7±0.15	22.30 (27)	33.80 (32)	8.26 (6.42)	9.35 (8.02)	72.64 (64.32)	15.02 (10.61)
Thousand grain weight (g)	28.2±0.80	29.01±0.31	18.60 (22.6)	38.15 (36.3)	19.02 (15.40)	24.03 (17.78)	62.69 (74.20)	31.02 (27.32)
Grain yield (t ha^{-1})	6.17±0.14	4.91±0.42	4.15 (2.73)	8.16 (6.7)	4.61 (6.08)	6.31 (8.80)	53.39 (47.78)	6.95 (8.66)
Biological yield (t ha^{-1})	13.58±0.18	13.4±0.20	9.25 (9.25)	16.24 (16.97)	3.20 (3.42)	4.51 (4.20)	50.34 (66.45)	4.68 (5.75)
Harvest index (%)	39.05±1.01	34.29±0.69	26.09 (23.30)	51.70 (46.90)	14.60 (14.17)	18.34 (20.97)	63.35 (45.65)	23.93 (19.72)
Day to heading (d)	181.05±0.42	181.7±5.8	174.8 (175.8)	188.50 (188.8)	2.16 (2.02)	2.29 (2.11)	88.73 (91.73)	4.18 (3.98)
Proline (U g^{-1} FW)	60.60±2.84	75.90±3.52	33.50 (44.30)	92.30 (127.00)	4.38 (4.27)	7.76 (5.98)	31.80 (50.92)	5.08 (6.27)
Protein	0.13±0.00	0.13±0.01	0.11 (0.11)	0.15 (0.15)	13.32 (17.2)	24.33 (42.2)	30.00 (16.67)	3.38 (1.09)
POD (U g^{-1} FW)	78.40±1.76	76.30±1.55	57.80 (60.20)	97.40 (93.50)	2.26 (2.74)	3.80 (4.34)	35.13 (39.98)	2.76 (3.57)
APX (U g^{-1} FW)	907.90±27.53	819.40±5.93	804.70 (705.40)	976.90 (873.50)	2.81 (3.38)	3.15 (3.85)	69.09 (67.05)	4.82 (5.32)
CAT (U g^{-1} FW)	48.90±0.37	55.40±0.79	32.60 (42.05)	53.10 (61.30)	1.75 (3.99)	3.58 (7.59)	19.16 (22.26)	1.57 (3.48)
SOD (U g^{-1} FW)	662.30±31.94	666.60±30.12	358.20 (334.10)	1089.70 (1067.40)	1.30 (1.52)	3.43 (3.28)	14.44 (21.60)	1.09 (1.46)

4. CONCLUSIONS

Results showed that mean grain yield over two growing seasons and under drought stress conditions was higher for the landraces KC4557, KC4862, KC3891, KC4495, KC4633, KC4537, KC2177, KC4619, KC3885, KC4618, KC4512 and KC4551. Multivariate analysis was used to assess the interrelationships of traits and the structure of landraces population under normal irrigation and drought stress trials in two years. Results of factor analysis indicated that 7 factors under irrigated conditions and 6 factors under drought stress respectively explained 80 and 75% of cryptic relationship behind the variation of traits. The first factor mainly defined by grain yield, grain number per spike, thousand grain weight, harvest index and number of spikelet per spike under fully irrigated conditions while factor 2 was related to the size of crop. Under drought, the first factor had the highest eigen value and explained 30% of total variations. Grain yield and its components had the highest loading coefficients in the first factor and this factor can be defined as yield components factor. Coefficients of factor 2 showed that this factor is a contrast between antioxidant enzymes and morphological traits. Antioxidants were not screenable parameters for drought tolerant genotypes because their accumulation increased in all genotypes in response to drought and had not strong correlation with grain yield. Results of stepwise analysis showed that thousand grain weight, biological yield, harvest index and spike length entered to the model as significant traits that highly contributed to grain yield variations under fully irrigated conditions. Under drought, thousand grain weight, biological yield, harvest index and grain number per spike entered to the model of grain yield and other traits excluded from the model as they were not significant.

Grain yield had higher correlations with thousand grain weight, harvest index, spikelet per spike under both fully irrigated and drought stress conditions. Canopy temperature had significantly negative correlations with grain yield under drought stress in both growing seasons. Narrow angles between the vectors of thousand grain weight, grain number per spike, spikelet per spike and harvest index indicate that these traits were highly associated with drought tolerance. Canonical analysis showed that grain yield and number and thousand grain weight had positive effects on drought tolerance while canopy temperature had negative effects on grain yield. According to the first antioxidant canonical variable, SOD, proline and CAT had higher contribution to drought tolerance compared to other enzymatic antioxidants. Cross correlations between opposite canonical variables and original variables indicated that protein content was decreased as a consequence of increased grain yield and antioxidant accumulations. Results of canonical analysis revealed that POD, CAT and APX as enzymatic antioxidants had negative cross associations with agronomic canonical variable under drought stress conditions which shows reduced grain yield as a consequence of antioxidant accumulation. Cluster analysis indicated that cluster 4 had lowest day to heading while cluster 5 had highest grain yield mean (5.3 t ha^{-1}) under drought. Therefore, these clusters were extremes for early maturity and grain yield and hybridization between the genotypes of clusters 4 and 5 is recommended for the introgression of grain yield and early maturity in a single cultivar as an approach for drought tolerance improvement. Thousand grain weight and harvest index had highest genetic advance predictors and it can be considered as selection index for drought tolerance improvement.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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