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Exploring genetic bases of heat stress tolerance and quality traits in wheat and their interplay

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Abstract

Wheat isprimaryfood grain produced in Pakistan. Fluctuation in environmental conditions among and within growing season is serious constraints influencing qualitative and quantitative wheat yield. Wheat is sensitie to heat stress. Terminal heat stress is one of the major causes of truncated productivity in the late sown conditions in Pakistan. The study was aimed for finding important traits and best genotypes in relation to heat stress. Thirty genotypes were sown at two different sowing dates following RCBD and replicated thrice. Analysis of variance showed significant differences among genotypes for all studied traits. Zeleny and gluten depicted high broad sense heritability coupled with high genetic advance indicating the presence of additive gene effect. Most of the studied traits showed moderate heritability. Biplot analysis showed that G14 and G20 had high 1000-grain weight and harvest index while G24, G25 and G10 present good quality parameters under heat stress.

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Introduction

Wheat is used as a staple food in Pakistan its growth and yield is badly affected by heat stress (Siebert and Ewert, 2014). Increasing temperature in many parts of the world is one of the major limiting factors for better production of wheat (Ortiz *et al.*, 2012). Its production is severely threatened in many countries by heat stress especially during reproductive and grain-filling stages. It is predicted that increased temperature will reduce wheat production by 20-30% in developing countries (Lobell *et al.*, 2008).

Changes in cropping pattern due to delayed sowing reduce grain yield, tillering period and high risk of hot weather during grain filling (Khokhar *et al.*, 2010). Heat stress during post flowering and early grain filling results in increased rate of leaf senescence (Telfer *et al.*, 2013). Loss of chlorophyll reduce incorporation of current carbon into grains so stay green genotypes can effectively retain grain filling under raised temperature (Farooq *et al.*, 2011).

Delayed sowing had maximum detrimental effect on 1000-grain weight (Hakim *et al.*, 2012). Canopy temperature is negatively correlated with yield under heat stress and cool canopy during grain filling period in wheat is an important physiological principle for high temperature stress tolerance (Mason and Singh, 2014).

Bilge *et al.* (2008) reported that canopy temperature depression is positively correlated to grain yield. Delayed sowing caused reduction in days to heading and plant height (Hakim *et al.*, 2012; Laghari *et al.*, 2012). This is may be due to reduction of life cycle caused by terminal heat stress associated with late sowing. Late plantation also caused reduction in biomass, harvest index and grain yield (Khan *et al.*, 2007; Singh *et al.*, 2011; Suleiman *et al.*, 2014).

In context of quality late planting improved bread quality and protein contents (Abdullah *et al.*, 2007). The investigations of Yadava and Singh (2003) showed that crude protein contents increased with delayed sowing. Heat stress during grain filling stage reduces starch deposition which increase protein concentration by letting more protein per unit of starch and enhancing grain protein contents (Gooding *et al.*, 2003). By increasing temperature starch biosynthesis and deposition is decreased which reduce grain dry matter. Although carbon and nitrogen daily flow into grains increases with increase in temperature but carbon flow decreases per degree day. In this way temperature affects grain size more than grain nitrogen quantity (Daniel and Triboi, 2000). However, heat stress increase grain protein content (Hakim *et al.*, 2012) but functionality of proteins is decreased considerably as it effects the development of compound protein aggregate which is required for positive dough mixing properties (Corbellini *et al.*, 1997). Heat stress limits glutenin synthesis and increase gliadins synthesis (Majoul *et al.*, 2003). As the protein content increase the zeleny index decreased and deterioration of gluten protein occurs (Balla *et al.*, 2011). Heat stress limits grain yield so, total protein content of crop reduced (Castro *et al.*, 2007). Maximum increase in grain protein content occurs when plants exposed to heat stress during early grain filling phase (Castro *et al.*, 2007). The present study was designed with the following objectives *i.e.* characterization of wheat genotypes under normal and late sowing conditions, checking potential of different wheat varieties under heat stress and assessment of contribution of different traits towards heat stress tolerance to develop heat tolerant varieties of wheat.

Material and methods

The present study was conducted in the experimental area of Wheat Research Institute, Ayub Agricultural Research Institute, and Faisalabad, Pakistan. Treatments comprised of two dates of sowing, normal was sown in 12-11-2017 and late sowing was done on fourth 15-12-2017. Experimental material was comprised of following thirty genotypes including five standard varieties:

These thirty genotypes were sown following randomized complete block design with three replications (Table 1). The gross plot size was kept six rows (length of row was six meter each, whereas distance between rows 30 cm) while plot size of six rows of five meter each was harvested to record data for grain yield and biomass. Normal agronomic and cultural practices were applied to the experiment throughout the growing season.

Measurement of traits

A plant was judged to be at heading when half of the spike is emerged from flag leaf. Normalized difference vegetation index (NDVI) was calculated by using Green Seeker. Canopy temperature was measured using hand-held digital Infrared thermometer LT300.

Plant height was measured with meter rod. Test weight was measured using test weight apparatus. Protein, starch, gluten and zeleny contents were recorded by using Bruins Instruments Omeganalyzer G.

Statistical analysis

Analysis of variance (Steel *et al.*, 1997) was conducted for each character. Heritability in broad sense was estimated according to Burton and Devane (1953).

Genetic advance was measured according to Allard's (1960). The mean data was analyzed for principal component analysis through R 3.5.2 software.

Results and discussion

Genetic diversity is a prerequisite for success in any breeding program. Analysis of variance depicted significant differences among genotypes for all studied traits (Table 2). The presence of wide range of diversity offers a huge scope for improvement through effective selection of desirable genotypes.

Table 1. List of wheat genotypes used in the experiment.

Sr. No.	Parentage
G1	ATTILA*2/PBW65*2/4/BOW/NKT//CBRD/3/ CBRD
G2	AUQAB 2000/CHAM 6
G3	AUQAB-2000/BAVICORA M-92
G4	CHEN/AEGILOPS SQUARROSA (TAUS)//BCN/3/BAV92/4/BERKUT
G5	CHENAB2000/INQILAB-91
G6	CHENAB2000/SHALIMAR88
G7	CHIL/2*STAR/4/BOW/CROW//BUC/PVN/3/2*VEE#10/5/UQAB 2000
G8	CNO79//PF70354/MUS/3/PASTOR/4/BAV92*2/5/FH6-1-7
G9	F 60314.76/ MRL// CNO 79/3/ LUCO-M/4/HEI/ 3* CNO 79// 2* SERI/5/ KAUZ// BOW/NKT
G10	FAISALABAD-08
G11	FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP// KAUZ/5/PFAU/WEAVER//BRAMBLING
G12	HUW234/5/CHIL/2*STAR/4/BOW/CROW// BUC/PVN/3/2*VEE#10
G13	INQ.91/FRET.2
G14	KACHU#1/4/CROC_1/AE.SQUARROSA (205) //KAUZ/...
G15	KFA/2*KACHU
G16	KOHISTAN 97/4/PASTOR/3/VEE # 5 //DOVE/BUC
G17	KOHISTAN-97/SOVA
G18	LASANI-08
G19	Millat-11
G20	NAC/TH.AC//3*PVN/3/MIRLO/BUC/4/2* PASTOR/5/...
G21	PB 96/V 87094// MH.97/3/UQAB.2000
G22	PRL/2*PASTOR//INQ.91
G23	Punjab-11
G24	SEHER-06
G25	SHALIMAR. 88/ 87094/ MH. 97/3/ LU.26/HD.2179//2*INQ. 91
G26	SHALIMAR. 88/ 87094/ MH. 97/3/LU. 26 /HD.2179//2*INQ. 91
G27	UQAB2000/CHENAB2000
G28	Uqab-2000/Punjab-85
G29	WBLL1*2/KURUKU//HEILO
G30	WEEBLI/CHENAB2000

Heritability and Genetic advance

The heritability can be demonstrated as proportion of total variance resulted due to genetic cause and factor that expressed degree of resemblance for different traits between parents and off springs. Heritability in broad sense also depicts relative achievement of selection. However, it is necessary to state that estimate of heritability is more effective for population from which worked out as they are not constant. Under normal conditions zeleny and gluten showed high heritability (≥60%) while NDVI at vegetative stage and 1000-grain weight showed low heritability (<30%).

Table 2. Mean squares values for all the studied traits under normal and heat stress conditions.

Parameters	Normal	Heat stress
Days to heading	18.458 **	7.597 **
Normalized difference vegetation index at vegetative stage	0.0008*	8.310×10^{-4} **
Canopy temperature at vegetative stage	0.817 **	1.024 *
Normalized difference vegetation index at reproductive stage	0.0018 **	0.002 **
Canopy temperature at reproductive stage	1.137 **	2.580 **
Plant height	58.149 **	105.502 **
1000-grain weight	16.412 *	22.9874 **
Grain yield	863711 **	501791 **
Biomass	3449517 **	1102482 *
Harvest index	0.0012 **	0.00221 **
Test weight	5.359 **	10.412 **
Protein	3.136 **	1.128 **
Starch	2.435 **	1.574 **
Gluten	53.973 **	15.410 **
Zeleny	119.459 **	126.228**

"**" significant at 1% probability level, "*" significant at 5% probability level.

Remaining parameters expressed moderate heritability 30 to 60%. However, Ali *et al.*, 2008; Jamali *et al.*, (2008); Yagdi and Sozen (2009); Din *et al.*, 2010 and Choudhary *et al.*, 2015 found high heritability for plant height, 1000-grain weight, harvest index and grain yield. Substantial improvement may be obtained through selection for these traits. For delayed sowing, canopy temperature at vegetative stage, biomass and starch had low heritability (25.25, 22.80 and 27.68%, respectively). However, canopy temperature at reproductive stage (72.86%) and zeleny (88.03%) showed high heritability while, all other traits depicted moderate heritability. Lopes and Reynolds (2012), found low to moderate heritability of canopy temperature at grain filling stage. Gutierrez *et al.,* 2010 reported that canopy temperature had strong correlation with grain yield under heat stress environment. High magnitude of heritability exhibited less environmental effect on these traits. While, parameters showed low heritability values were considered to be highly influenced by environmental effect and selection will not be useful in early generations. Canopy temperature and NDVI can be used as indirect selection tool for higher grain yield (Baber *et al.*, 2006).

Genetic advance expressed the potential gains which could be anticipated from the parameters considered over both environments. It demonstrates the gain that can be attained for the character in next generation. It is not compulsory that traits having high heritability must have high genetic advance. Under normal environment gluten and zeleny showed high genetic advance while grain yield exhibited moderate genetic advance (Table 3). However, grain yield and zeleny expressed moderate genetic advance ranged between 10 to 20% under heat stress conditions. Traits such as zeleny and grain yield depicted high to moderate heritability coupled with high genetic advance exhibited additive gene action thus selection is suggested for improvement. Traits with low heritability and low genetic advance depicted dominance gene action and thus heterosis breeding is recommended for traits improvement.

Principal component analysis under normal growing conditions

Principal component analysis (PCA) was performed to obtain more reliable information on how to identify groups of genotypes with desirable yield traits for planning breeding programmes. It also helps to differentiate significant relationship between traits.

Table 3. Estimates of heritability and genetic advance for various traits of wheat.

Parameter	Normal		Heat stress	
	h^2 (B.S)	GA (%)	h^2 (B.S)	GA (%)
Heading	52.46	2.84	42.84	1.87
NDVI.1	20.37	1.06	41.30	2.03
CT.1	52.98	3.36	25.25	1.77
NDVI.2	48.89	3.65	49.46	3.78
CT.2	39.93	2.61	72.86	5.29
PH	32.39	3.06	51.92	7.12
TGW	24.18	4.20	40.38	8.21
GY	53.14	12.52	53.48	12.58
Biomass	45.15	7.98	22.80	3.74
HI	32.39	4.27	51.38	7.60
TW	50.39	2.14	47.05	2.85
Protein	44.41	8.15	37.88	4.20
Starch	42.39	1.60	27.68	0.89
Gluten	70.92	28.14	36.44	9.21
Zeleny	80.28	20.39	88.03	19.50

(Heading= Days to heading, NDVI.1= Normalized difference vegetation index at vegetative stage, CT.1= Canopy temperature at vegetative stage, NDVI.2= Normalized difference vegetation index at reproductive stage, CT.2= Canopy temperature at reproductive stage, PH= Plant height, TGW= 1000-grain weight, GY= Grain yield, HI= Harvest index, TW= Test weight, h^2 (B.S) = Heritability in broad sense, GA%= Genetic advance percentage).

Under normal growing conditions first five PCs out of total fifteen had eigen value >1.

These five components contributed 75.6% of the total variability due to wheat genotypes evaluated for various morpho-physiological and quality traits (Table 4).

First component contributed 32.9% towards the total variability. In the first PC protein, zeleny, gluten, 1000 grain weight, harvest index and grain yield had maximum loadings. These traits would be enough to distinguish group of genotypes (Table 5). In this dimension varieties had less value for 1000-grain weight and harvest index while higher values for protein, zeleny and gluten. The sign of the loading specifies the trend of the relationship between the component and the variable.

PC-II contributed 14.5% towards variability (Table 4) and most of the variations among genotypes were explained by biomass, NDVI.2, grain yield and heading (Table 5).

In this component varieties exhibited high values for biomass, NDVI.2, grain yield and heading.

Table 4. Eigen values and variance of principal component for 15 characters in 30 genotypes of *Triticum aestivum* L. under normal conditions.

Parameter	PC_1	PC_2	PC_3	PC_4	PC_5
Eigenvalue	4.9366	2.1788	1.6026	1.4566	1.1612
Variance	32.9	14.5	10.7	9.7	7.7
Cumulative	32.9	47.4	58.1	67.8	75.6

Table 5. Principal component for 15 characters in 30 genotypes of *Triticum aestium* L. under normal conditions.

Variables	PC1	PC2	PC3	PC4	PC5
Heading	0.203	0.373	0.169	0.061	0.372
NDVI.1	0.232	0.239	0.217	-0.533	-0.144
CT.1	-0.108	-0.201	0.051	-0.361	0.474
NDVI.2	0.220	0.439	-0.225	-0.176	0.044
CT.2	0.167	0.196	0.354	0.129	-0.456
PH	0.049	-0.124	0.381	0.381	-0.091
TGW	-0.306	-0.152	-0.187	0.299	0.067
GY	-0.313	0.425	-0.089	0.173	-0.018
Biomass	-0.201	0.512	-0.143	0.213	-0.061
HI	-0.375	0.099	0.051	0.008	0.059
TW	-0.064	0.172	-0.053	0.165	0.540
Protein	0.384	0.026	0.514	-0.155	0.283
Starch	-0.294	0.024	0.311	0.304	-0.072
Gluten	0.284	-0.088	-0.283	0.224	0.085
Zeleny	0.352	-0.049	0.302	0.175	-0.041

(Heading= Days to heading, NDVI.1= Normalized difference vegetation index at vegetative stage, CT.1= Canopy temperature at vegetative stage, NDVI.2= Normalized difference vegetation index at reproductive stage, CT.2= Canopy temperature at reproductive stage, PH= Plant height, TGW= 1000-grain weight, GY= Grain yield, HI= Harvest index, TW= Test weight).

Third PC explained 10.7% of the total variation (Table 4). This PC explained by differences among genotypes due to maximum loading of plant height, protein and starch. Third PC represented positive effect for protein, plant height, and starch which means varieties in this component had high values for these characters (Table 5).

The fourth PC described 9.7% of total differences among genotypes (Table 4). This PC showed maximum loading for plant height and NDVI. 1 (Table 5). Varieties of this component had less value for NDVI.1 and high for plant height means both traits have negative correlation.

PC-V contributed only 7.7% towards total variability (Table 4). Four traits i.e. days to heading, CT.1, CT.2 and test weight had maximum loadings in this component (Table 5). The varieties of this component have less value for CT.2 and high value for test weight. Similar studies were carried out by Golparvar*et al.*, 2003a, b; Leilah and Al-Khateeb, 2005 and Beheshtizadeh *et al.,* 2013.

Biplot

A principal component biplot (Fig. 1) showed that variables were super imposed on the plot as vectors. Distance of each with respect to PC1 and PC2 showed the contribution of this variable in the variation of germplasm.

Table 6. Eigen values and variance of principal component for 15 characters in 30 genotypes of *Triticum aestivum* L. under heat stress.

Parameter	PC_1	PC_2	PC_3	PC_4	PC_5
Eigenvalue	4.3627	2.1911	1.7775	1.5885	1.1782
Variance%	29.1	14.6	11.8	10.6	7.9
Cumulative%	29.1	43.7	55.5	66.1	74.0

Table 7. Principal component for 15 characters in 30 genotypes of *Triticum aestium* L. under heat stress conditions.

Variables	PC1	PC2	PC3	PC4	PC5
Heading	0.065	0.091	-0.479	-0.104	-0.102
NDVI.1	0.055	0.499	-0.002	-0.307	-0.291
CT.1	0.013	0.257	0.355	0.394	-0.462
NDVI.2	0.086	0.550	-0.015	-0.253	-0.209
CT.2	-0.088	0.216	0.344	0.186	0.120
PH	0.024	0.227	-0.500	0.429	-0.116
TGW	0.305	-0.127	0.273	0.301	-0.273
GY	0.449	-0.069	-0.122	-0.006	-0.033
Biomass	0.369	0.081	-0.262	-0.023	0.096
HI	0.398	-0.188	0.045	0.023	-0.161
TW	0.360	-0.215	-0.034	0.030	-0.210
Protein	-0.249	-0.284	-0.178	-0.054	-0.502
Starch	0.371	0.011	0.109	-0.085	0.180
Gluten	-0.097	-0.280	0.088	-0.471	-0.401
Zeleny	-0.223	-0.090	-0.251	0.365	-0.136

(Heading= Days to heading, NDVI.1= Normalized difference vegetation index at vegetative stage, CT.1= Canopy temperature at vegetative stage, NDVI.2= Normalized difference vegetation index at reproductive stage, CT.2= Canopy temperature at reproductive stage, PH= Plant height, TGW= 1000-grain weight, GY= Grain yield, HI= Harvest index, TW= Test weight).

The first component can be termed as quality of grains as it containing genotypes that have high values of these parameters. Similarly second PC can be named as yield potential and it represented the high yielding genotypes from less yielding genotypes. Genotypes number G12, G14, G20 and G26 have maximum value for grain yield, biomass, harvest index and starch under normal planting (Fig 1).

While, genotype G13, G15, G23 and G24 possessing maximum values for protein, gluten and zeleny but having less yield and 1000 grain weight. The genotype number 16 have high value for NDVI.1, NDVI.2, CT.1 and took more days to heading whereas, other genotypes positioned near origin behave neutrally for all studied traits.

Principal component analysis under heat stress conditions

Under heat stress conditions first five PCs out of total Fifteen had eigen value >1. These five PC contributed 74% of total diversity among studied genotypes.

Maximum variability of 29.1% was contributed by PC1 (Table 6). In first PC grain yield, harvest index, starch and biomass have maximum loadings. These traits would be enough to distinguish group of genotypes (Table 7). This PC represented positive effect for grain yield, harvest index, starch and biomass which means varieties in this component had high values for these traits.

PC-II contributed 14.6% of total variability among genotypes (Table 6). PC-II had maximum loading for NDVI.1, NDVI.2, protein and gluten (Table 7). The varieties of this component had high values of NDVI.1 and NDVI.2 but less protein and starch contents.

Third PC explained 11.8% of total variability (Table 6). This PC explained by differences among genotypes due to maximum loadings of heading, plant height, CT.1, and CT.2 (Table 7). In this component varieties have short stature and took less days to head emergence but have high canopy temperature at reproductive and vegetative stage.

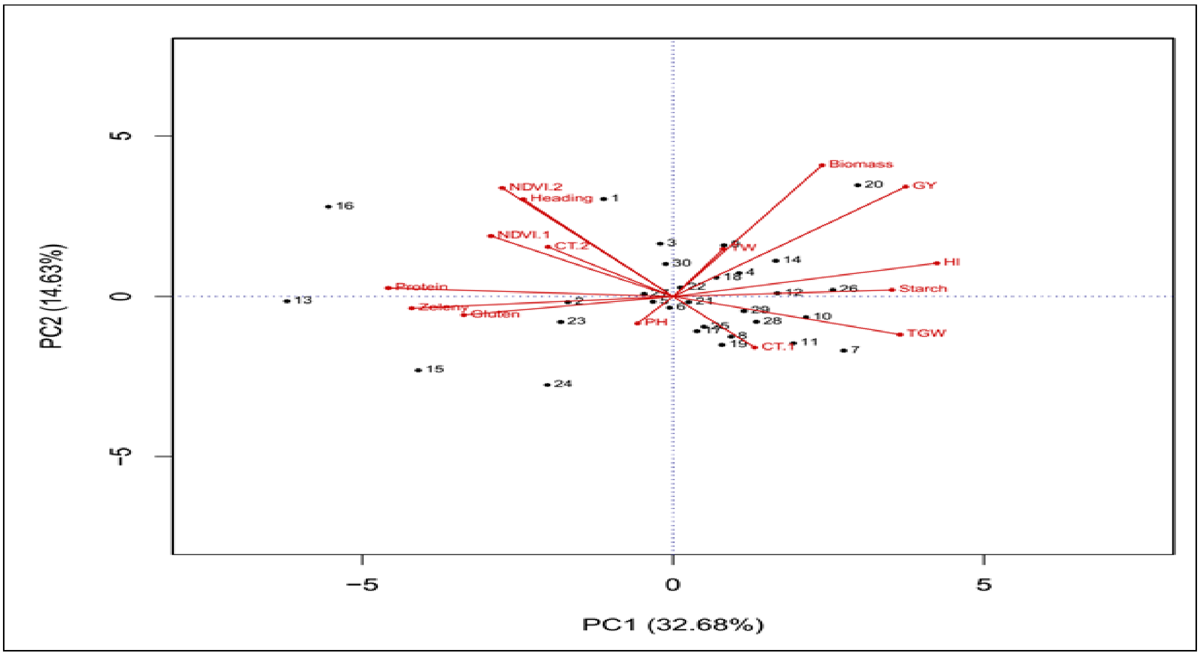

Fig 1. Biplot graphical display of the studied traits in bread wheat cultivars under normal conditions.

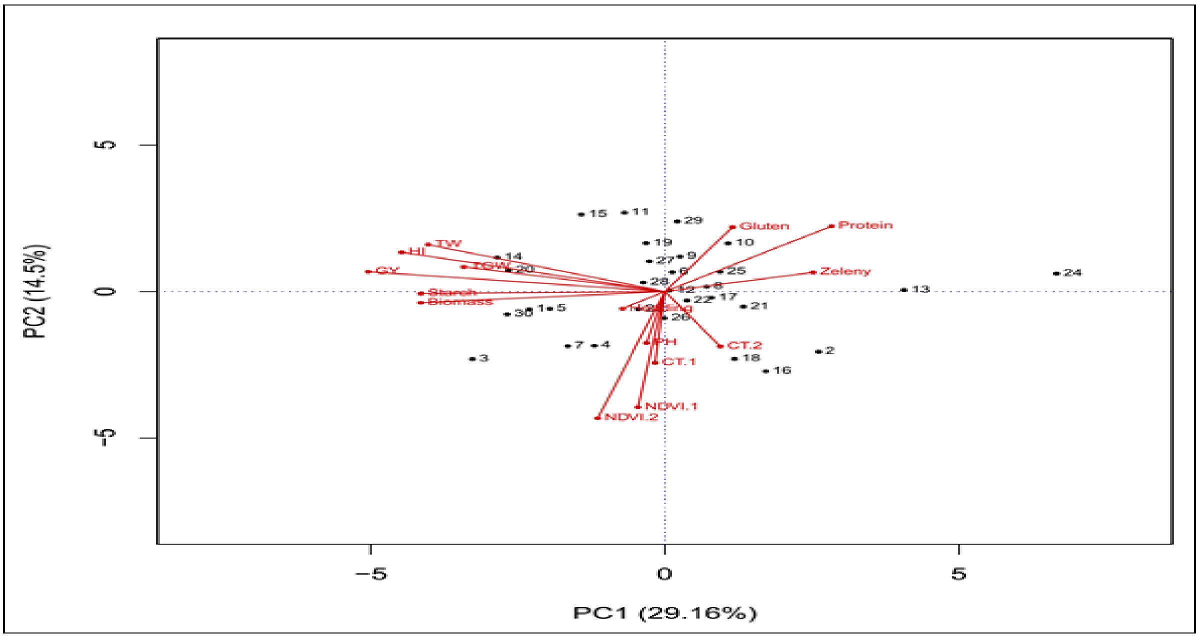

Fig. 2. Biplot graphical display of the studied traits in bread wheat cultivars under heat stress conditions.

The fourth PC contributed 10.6% of total variability (Table 6). This PC showed maximum loadings for gluten, plant height, CT.1 and zeleny (Table 7). Varieties of this component have less gluten percentage and have high values of CT.1, plant height and zeleny. PC-V shared only 7.9 % of total variability (Table 6). Three traits i.e. CT.1, gluten and protein have maximum loadings. This component represented negative effect for CT.1, gluten and protein which means this component consisted of varieties having less value for these traits and negative correlation among each other (Table 7).

Biplot

Under heat stress conditions, PC1 considered as yield potential and related parameters due to high positive value of these traits. This PC represents the high and low yielding genotypes from the standard genotypes. Genotypes 11, 14, 15, 20 and 30 are more suitable genotypes for heat stress conditions due to high yield and heat tolerance. As they had high values for grain yield, harvest index, starch, test weight and biomass (Fig. 2).

Genotypes 10, 13 and 24 had high protein, gluten and zeleny contents. Genotypes 16 and 18 were poorest as they had higher canopy temperatures at vegetative as well as reproductive stage (Fig. 2).

Conclusion

It has been concluded from above discussion that experimental material had highly significant genetic variability for all the studied traits.

It is suggested that phenotypic selection would be more beneficial for characters with high values of heritability accompanied by high genetic advance.

High heritability accompanied by low to moderate genetic advance for days to heading, canopy temperature at vegetative stage, grain yield and test weight under normal conditions while for plant height, grain yield and harvest index under heat stress conditions, may be attributed to non-additive gene action and may further be improved through hybridization. Genotypes 11, 14, 15, 20 and 30 are more suitable genotypes for heat stress conditions due to high yield and heat tolerance.

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