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EVALUATION OF YIELD COMPONENTS AND HEAT SUSCEPTIBILITY OF PAKISTANI WHEAT (*TRITICUM AESTIVUM* L.) GERMPLASM SUBJECTED TO INDUCED TERMINAL HEAT STRESS

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Abstract

A set of 205 Pakistani wheat genotypes was evaluated to access the endogenous potential for heat tolerance and agro-morphological genetic diversity in wheat population subjected to induced terminal heat stress. About 10 out of 21 agro-morphological traits, showed higher coefficient of variation (CV%) in comparison to control, which was directly associated with extent of variability in subjected population. Various traits showed a strong positive correlation in case of treatment and control. First two axes of bi-plot, F1 and F2 of principal component, PC1 as well as PC2 explained 17% and 12% for year 2017-18, 16% and 15% for year 2018-19, as well as 14% and 12% variability under control conditions respectively. Total of 8 genotypes showed heat susceptibility index (HSI≤ 0.5), hence designated as highly heat tolerant (HHT) genotypes. These genotypes also showed ≤ 20% cumulative reduction in 1000-kernel weight in comparison to control. On the bases of green leaf retention (GLR) scale, population was classified into 4 categories i.e., 11, 11% resistant (R), 48, 41% moderately resistant (MR), 22, 41% moderately susceptible (MS) and 19, 7% susceptible (S) during year 2017-18 and 2018-19 respectively. This data will be used to perform genome wide association studies (GWAS) to identify QTLs associated with heat stress tolerance.

Key words: Genetic diversity, Principal component analysis, Correlation analysis, Heat susceptibility index (HSI). Thousand kernel weight reduction (1000 KWR), Green leaf retention (GLR).

Introduction

Wheat (*Triticum aestivum* L.) or spring wheat is a principal macro as well as micro nutrient bearing caloric source of food for 2.5 billion (>40%) (Arzani & Ashraf, 2017) people, in 89 countries of this planet. It adds up to 20% protein and energy in daily human diet (Anon., 2017; Fischer *et al*., 2014; Gupta *et al*., 2008; Hawkesford *et al*., 2013). Wheat single handedly producing 19% edible grains for world cereal market. Merely wheat is providing 55% of the carbohydrates needed for human food. Wheat is cultivated over 224 million hectares annually which accounts, about 19% of the world arable farmlands. Global annual wheat production of 765 million metric tons (MMT) was expected for the cropping year 2018-19 (Anon., 2019). Somehow are the others 68% of the world wheat stocks are consumed by the human beings and rest of the 32% are reserved for livestock's and other miscellaneous uses.

Wheat demand will increase up to 70%, meanwhile, the mean temperature in South Asian region will also rise up to 4°C by the year 2050 (Borlaug & Dowswell, 2003; Loisel *et al*., 2021; Stratonovitch & Semenov, 2015). World annual wheat production is increasing with a pace of 0.5-1% per annum (Crespo-Herrera *et al*., 2017), but such visual statistics of crop production are not sufficient to meet the challenges food security. It is upraised that nearly 2% annual projection in genetic gains are direly of global needed to ensure the world food security (Chattopadhyay, 2010; Douglas, 2009; Garcia *et al*., 2019; Gill *et al*., 2004; Maulana *et al*., 2018; Tanaka *et al*., 2015). Global food security is turning into a snowballing challenge due to continuous increase in population and a swift decrease in arable wheat farmlands. It was estimated that about 1ha agricultural land is camouflaged in every 7.7 seconds due to prompt climate change, industrialization and urbanization (http://irri.org/). Shortage of water, input resources, climate uncertainties and various other biotic and abiotic factor continuously reducing crop production as well as averting sustainable world food security challenge (Yang *et al*., 2012).

Climate variation executes a bunch of biotic and abiotic anomalies on wheat crop but unexpected heat wave are extremely deleterious for wheat production in indo-genetic plains of South Asia. A brief episode of high temperature from anthesis to grain filling until late maturity is termed as terminal heat stress that possess a divers effect on wheat production (Wahid *et al*., 2007). In South Asia such as India, Pakistan and Bangladesh roughly 60% wheat is late sown due to delayed harvesting of preceding Kherif season crops. It's obvious from the fact that about 58% of the global wheat arable farmland are badly affected by the heat stress (Crespo-Herrera *et al*., 2017; Kosina *et al*., 2007). In previous decade various studies to gauge the impact of climate variability on bread wheat production were performed in India and Bangladesh (Lobell *et al*., 2012; Sarker *et al*, 2012; Mondal *et al*., 2013; Krupnik *et al*., 2015; Krupnik *et al*., 2015).

In Pakistan~ 70% of the rural farmer community directly or indirectly derive their livings from agriculture farms. Pakistan is 7th largest wheat producers of the world and 3rd biggest wheat producer of Asia pacific. Bread wheat individually providing 75% of the countries caloric need (Timsina & Connor, 2001). *Triticum aestivum* L. is an important cash crop for Pakistani farmer, which solely accounts 8.7% value addition in agriculture sector and 1.7% in countries gross domestic product (GDP). During previous Rabi season wheat was cultivated on an area of 8,825 thousand hectares. In Pakistan, wheat production reaches to 24, 946 million tons with 2.5% increase from the last fiscal year 2018-19 due to increase in cultivation area and healthy rain at grain formation (Anon., 2018-19). A swift increase in Pakistani population at a rate of 2.4% per annum augment a stern challenge of sustainable food security in front of country think tanks as well as for agricultural scientific community of the county. Pakistan is turning to be the most vulnerable country regarding prompt catastrophic climate changes in South Asia (Nasim *et al*., 2016; Crimp *et al*., 2019). Significant temperature fluctuation was observed over 30 sites in pre-monsoon season as well as during the month of March in Pakistan (Iqbal *et al*., 2014). According to (Crimp *et al*., 2019) 0. 10 to 0.18°C increase in global temperature was augmented.

Moreover 0.57 to 1.31 t/ha^{-1} yield variability in rice wheat cropping system was observed in national production of Pakistan during first decade of the current millennium (Anon., 2019). There is no apprehension to say that climate variation is a solitary significant yield determining factor elucidating 30-50% world crop yield fluctuation (Frieler *et al*., 2017; Ray *et al*., 2015; Zampieri *et al*., 2017) Merely 1°C temperature projection leads up to 4.1% to 6.4% yield fluctuation in bread wheat (Liu *et al*., 2016b) Climate change i.e. temperature variation does not only squeeze the crop cycle but also accelerate the crop development rate by escalating the frequency of unexpected heat episodes (Asseng *et al*., 2015). A single event of suboptimal temperature during anthesis, grain filling duration or till crop maturity (terminal heat stress) in cereal crops considerably reduce crop productivity (Rezaei *et al*., 2015; Talukder *et al*., 2014). Terminal heat stress harnesses a significant negative effect on all post-anthesis traits such as, squat crop cycle, early heading, early maturity, reduced number of spikelet per spike, pollen sterility, loss of chlorophyll, accelerated photorespiration, reduced grain number and size of grain per spike, shrieked deformed seed, suboptimal flour quality as well as low yield (Ghaffari *et al*., 2015). Terminal heat stresses also possess significant impact at cellular level. The most promising is the excessive production of Reactive Oxygen Species (ROS) that leads toward rapid oxidation of lipids of cellular membrane as well as terminate activities of rubisco that ultimately decreases photosynthesis and cell stability by disrupting cells internal homeostasis (Hasanuzzaman *et al*., 2013). A number of non-distractive methods such as stay green or green leaf retention, heat susceptibility index and kernel weight reduction are commonly used for the screening of heat tolerance in wheat crop. Some possible method i.e. timely sowing, breeding for heat tolerant, early maturing varieties having improved grain filling rate and extended pollen viability could be employed for crop improvement in Pakistan as well as in other heat affected regions of the world (Mondal *et al*., 2013; Reynolds *et al*., 2016).

Evolution is a key player behind this colossal morphological and genetic diversity in natural flora and fauna on this planet, but this process is too slow to meet the current global needs. Continuous selfing in self-pollinated crops led toward narrow genetic bases as well as loss of genetic vigor in modern cultivars. Wide natural selection, introduction, genetic hybridization, induced mutagenesis and horizontal as well as vertical gene transfer are needs of the hour to widen the genetic bases of modern wheat varieties to meet the challenges of climate change and food security. Greater genetic variability directly associated with the chances to develop genotypes of ideal characteristic (Khan *et al*., 2003; Aycicek *et al*., 2006; Arya *et al*., 2013).

Genetic resources such as wild relatives, gene bank accessions, landraces, breeders advanced lines, induced and natural mutants were considered as an indispensable genetic resources for genetic diversity and crop improvement. A comprehensive phenotypic foreground and background dissection of agro morphological and yield related traits is a prerequisite to initiate any strategic trait based breeding program (Reynolds *et al*., 2009). In current era of genomics and molecular breeding phenotypic information and genetic diversity estimation is always a first step towards crop improvement. Now a days various advanced genomic techniques such as association mapping (AM), genome selection (GS), and genome wide association studies (GWAS) also require effective, high throughput, phonemic information for strategic trait based crop improvement programs. Comprehensive phenotypic information is necessary for the selection of parents for trait based crossing, developing conceptual breeding models and for making authentic breeding decisions regarding genetic gains in crops (Reynolds & Langridge, 2016). Physiological trait (PTs) information of multi-location trials is also important for the selection, introgression, and pyramiding of positive alleles in advanced genotypes to pace up crop production with demands (Richards & Lukacs, 2002). The current experiment was designed to dissect the genetic variability in agro-morphological and yield traits as well as to evaluate the endogenous potential of heat tolerance in Pakistani genotypes under induced terminal heat stress conditions.

Materials and Methods

Germplasm collection and experimental design: Healthy seeds of 205 local genotypes (pedigree of all genotypes against their CB# see Supplementary Table 1 at the end) were collected from the “wheat research institute” (WRI), ayoub agricultural research institute (AARI), Faisalabad. The field experimental trials with three replicates and separate control for each year were conducted at (Latitude = 31°-26' 29.4" N, Longitude = 73°-04'37.7" E) for two consecutive years (2017-18 and 2018-19) at university of agriculture Faisalabad (UAF) experimental area in randomized complete block design (RCBD). Two healthy wheat seeds were sown in each hole and single plant per hole was maintained at four leaf stage. Three rows of 1.5m length in each replication were grown by keeping plant to plant as well as row to row distance of 15.24 and 22.86 cm respectively. However 30.48 cm distance was maintained between genotypes. A separate control was also grown during each year in similar field layout and standard agronomic practices were applied. Flow chart of experiment from sowing to harvesting presented is in (Fig. 1).

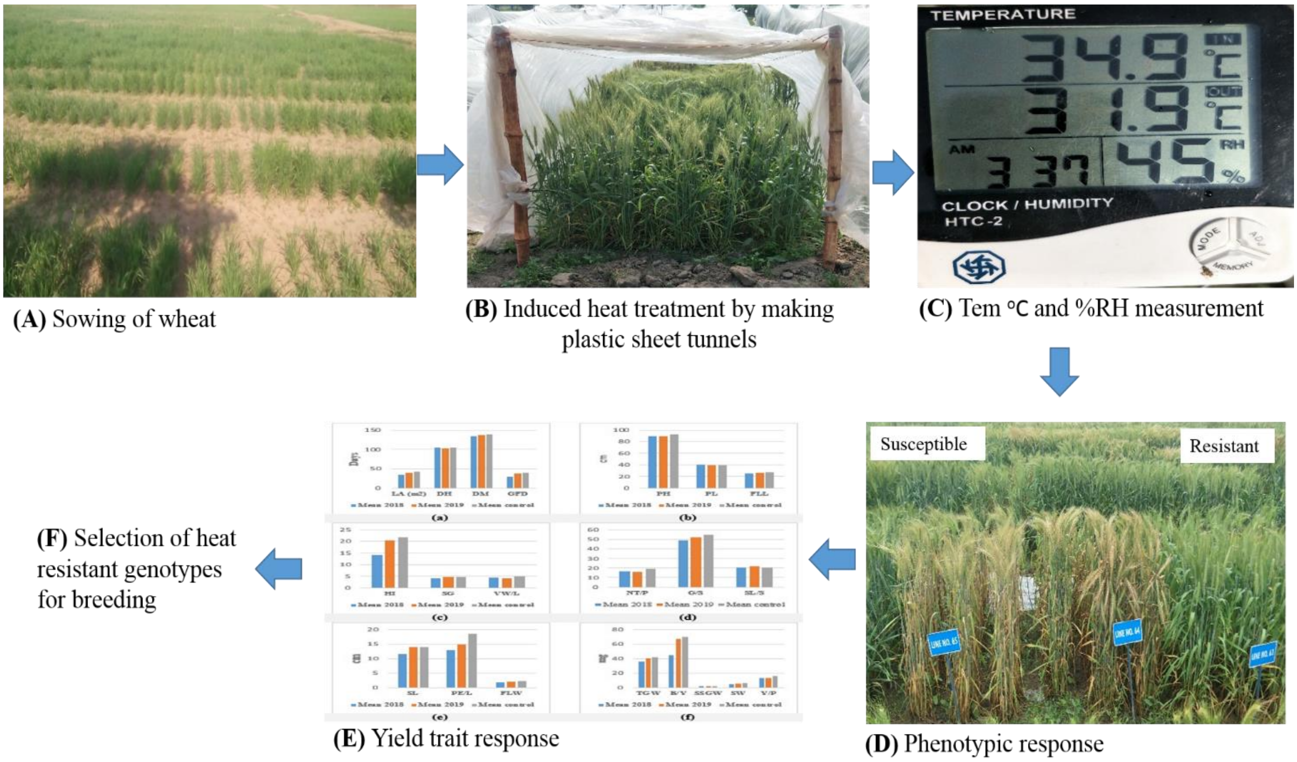


Fig. 1. Schematic flow chart of experiment, (A) sowing of wheat, (B) induced heat treatment, (C) measurement of temperature and relative humidity (%RH), (D) phenotypic response measurement, (E) comparison of yield related traits, (F) selection of heat tolerate genotypes for breeding.

Crop husbandry and artificial heat treatment: The wheat crop was sown at optimum recommended sowing time window in mid of month of November (during both growing season 2017-18 and 2018-19). Normal (Control) and treatment were raised under similar crop husbandry practices before application of heat stress. Recommended dose of nitrogen, phosphors and potash (N:P:K) at the rate of 46:34:25/kg/0.4047 hectare) was applied respectively. Three irrigations such as 1st, irrigation was applied after 25 to 30 days of sowing (tillering stage), 2nd irrigation was applied after 80 to 90 days at (botting stage) of crop and 3rd or final irrigation applied after 120 to 130 days at (milking stage) throughout crop life cycle. Flood irrigation method was used for irrigation purposes. Some specifically recommended weedicides and fungicides were applied for the control of weed and fungal diseases. Moreover five days after anthesis (DAA), temporary plastic sheath tunnels of (1.52m wide and 1.52m height) dimensions were raised to mimic terminal hot humid stress conditions from Zadoks growth stages GS 73 to GS 77). The impact of stress on a crop generally depends upon growth stage, longevity and intensity of stress applied. Keeping in view the recommendations of (Rahman *et al.*, 2009; Shahid *et al.*, 2017) minimal internal tunnel temperature of 32^{o}C was sustained as threshold for spring wheat. Such experimental conditions were opted for 10 days in both cropping years (2017-18 and 2018-19), following the methodolgy used by Farooq *et al.*, 2011; Balla *et al.*, 2011; Prasad & Djanaguiraman, 2014; Liu *et al.*, 2014 with modification. A portable field thermometer was used to record the temperature and humidity within tunnels. Induced heat stress conditions of both season treatment and control presented are in (Table 1).

Phenotypic response evaluation parameters: Three healthy plants, one from each replication were selected and tagged to collect data of agro morphological and yield parameters. However, 21 agro morphological and yield traits were selected for virtual screening of the germplasm. Plant height (PH), peduncle length (PL), peduncle extrusion length (PEL), spike length (SL), flag leaf width (FLW), flag leaf length (FLL), leaf area (LA) (were measured using the formula: Leaf area= FLLx FLW)x0.75)) by (Muller, 1991), spikelet per spike (SL/S), number of tillers per plant (NT/P), yield per plant (Y/P), harvesting index (HI) were calculated by applying the formula (Donald, 1968), HI= (YP/BY) x100)). Biological yield (BY), thousand grain weight (TGW), days to heading (DH), days to maturity (DM), grain filling duration (GFD), spike weight (SW), single spike grain weight (SSGW), grains per spike(G/S), abaxial flag leaf glaucousness or visual wax on leaf were calculated by using the 0-5 scale coined by the (Bennett *et al.*, 2012). Green leaf retention (GLR) or stay green (SG) was measured using 0-10 scale used (Li *et al.*, 2018). These selected traits were used as a yard stick to dissect the morphological diversity and response of the experimental population against induced terminal heat stress.

Table 1. Comparison of experimental conditions: (A) Temperature (Tem°C) and relative humidity (RH%) inside (Treatment) tunnel and outside of the tunnel (Control), during (Year 2017-18), (B) Comparison of experimental conditions temperature (Tem°C) and relative humidity (RH%) inside (Treatment) tunnel and outside the tunnel (Control), during (Year 2018-19).

Date	A			
	Tem. °C (inside Tunnel)	RH % (inside Tunnel)	Tem. °C (outside Tunnel)	RH % (outside Tunnel)
3/3/2017-18	32	65	22	68
3/4/2017-18	32	80	26	69
3/5/2017-18	34	66	26	70
3/6/2017-18	33	44	29	61
3/7/2017-18	32	56	29	70
3/8/2017-18	38	53	33	66
3/9/2017-18	37	52	33	66
3/10/2017-18	36	52	33	68
3/11/2017-18	41	67	35	65
3/12/2017-18	39	53	34	65
Date	**B**			
	Tem°C (inside Tunnel)	**RH% (inside Tunnel)**	**Tem°C (Outside Tunnel)**	**RH% (outside Tunnel)**
3/3/2018-19	32	65	24	80
3/4/2018-19	33	80	25	76
3/5/2018-19	34	66	24.5	61
3/6/2018-19	31	63	22	79
3/7/2018-19	33	56	25	71
3/8/2018-19	38	64	27	66
3/9/2018-19	33	52	25	62
3/10/2018-19	32	61	24	72
3/11/2018-19	32	67	22	70
3/12/2018-19	36	55	26	69

Table 2. GLR response measurement scale.

Scale	% of Green area	Groups	Response result
0-1	90-100% green	I (>0 and ≤1	Resistant
2-5	50-80%	II (≥2 and ≤5	Moderately Resistant
6-8	20-40%	III (≥6 and ≤8	Moderately Susceptible
9-10	0-10%	IV (≥9 and ≤10	Susceptible

Nondestructive methods for screening of heat tolerance: Three important methods i.e. Green Leaf Retention (GLR) is also known as Stay Green (SG), Kernel Weight Reduction (KWR) and Heat Susceptibility Index (HSI) were used to access the heat tolerance in this experiment.

Green leaf retention (GLR): It is the rating of wheat flag leaf retaining green color after induced heat treatment and measured using 0-10 scale (Li *et al*., 2018). For the sake of convenience GLR scale was divided in to four categories presented in (Table 2).

Kernel weight reduction (KWR): KWR is another method to access the response of wheat genotypes grown under stress conditions (Fokar *et al*., 1998). Difference in TGW between stressed and controlled condition was used to calculate the KWR using the following equation (Eq. 1).

$$KWR = 1 - kw/kwp \quad \text{(Eq. 1)}$$

Kw was 1000 kernel weight (stressed condition) and Kw_p showed thousand kernel weight (TKW) under (normal condition).

Heat susceptibility index (HSI): The HSI is another method used to screen wheat genotypes under heat stress conditions. It is commonly used to classify wheat genotypes with respect to the degree of heat tolerance. Fischer & Maurer, (1978) index was used to estimate the heat susceptibility index in wheat using the (Eq. 2).

$$HSI = \frac{1-Y/Yp}{D} \quad \text{(Eq. 2)}$$

Here Y is designated as mean of each genotype under stress conditions and Y_p is mean of each genotype under non stress (control) conditions. Stress intensity (D) is determined by using following expression $D = 1 - \frac{X}{Xp}$. Furthermore, X is denoted for mean of all genotypes under stress and Xp is mean of all genotypes under non stressed conditions. If the value of heat susceptibility index, HSI ≤ 0.5, HSI = 0.5-1.0 and HSI ≥ 1.0 represent the high heat tolerance, moderately heat tolerance and heat susceptible response respectively.

Table 3. Statistical summary of 21 agro-morphological traits of 205 Pakistani spring wheat genotypes for cropping year 2017-18, 2018-19 and control of respective season.

	Season 2017-18					Season 2018-19					Mean of control 2017-18 and 2018-19				
Traits	Range	Mean	Var. (n)	SD(n)	CV%	Range	Mean	Var. (n)	SD(n)	CV%	Range	Mean	Var. (n)	SD(n)	CV%
PH	62-112	89.56	90.09	9.49	10.60	62-110	89.53	88.87	9.43	10.53	65-115	92.56	90.09	9.49	10.25
PE/L	4-30	12.75	21.92	4.68	36.73*	5-23	14.79	13.23	3.64	24.60*	7-38	18.44	21.99	4.69	25.43*
PL	12-65	40.23	32.66	5.72	14.21	15-62	39.73	24.86	4.99	12.55	16-62	39.73	24.86	4.99	12.55
NT/P	3-30	16.82	35.74	5.98	35.54*	6-24	15.72	11.85	3.44	31.89*	5-32	18.82	35.74	5.98	31.77*
SL	7-16	11.52	3.35	1.83	15.89	10-19	14.02	2.78	1.67	11.90	10-19	14.02	2.78	1.67	11.90
SL/S	13-27	20.41	4.56	2.14	10.47	15-27	21.51	4.35	2.09	9.69	13-27	20.41	4.56	2.14	10.47
FLL	15-36	24.91	14.01	3.74	15.03	15-39	26.46	17.67	4.20	15.88	16-40	27.46	17.67	4.20	15.31
FLW	1.3-3	1.86	0.05	0.22	11.80	1.3-2.7	2.00	0.08	0.28	14.14	1.3-3	2.12	0.90	0.95	14.80
LA	17-57	34.54	60.21	7.76	22.47*	15.5-74	39.52	103.11	10.15	25.69*	17-77	42.10	109.8	10.44	24.81*
DH	90-114	104	12.74	3.57	3.44	88-112	102	12.70	3.56	3.50	90-114	104	12.72	3.57	3.43
DM	121-145	133	33.41	5.78	4.35	122-143	137	18.23	4.27	3.12	124-151	139	18.23	4.27	3.07
GFD	13-43	29	32.93	5.74	19.81	16-40	38	13.73	3.71	9.74	21-51	39	14.06	3.75	9.61
Y/P	3-25	13	17.53	4.19	32.35*	2.8-29	13	18.55	4.31	33.13*	4.3-31	15.5	18.55	4.31	29.70*
B/Y	14-107	44.70	332.74	18.24	40.81*	21-132	67.07	399.29	19.98	29.79*	24-135	69.65	389.2	19.73	28.33*
HI	4.7-88	14.08	324.15	18.00	52.82*	5.4-53	20.51	56.97	7.55	36.81*	6.9-53	21.83	54.20	7.36	33.72*
SSGW	.07-4.3	1.86	0.62	0.79	42.40*	.2-4.6	2.15	0.65	0.80	37.44*	2.3-4.7	2.25	0.65	0.80	35.77*
SW	1.7-7	4.42	1.15	1.07	24.31	3-8.6	5.69	1.15	1.07	18.88	3.7-9.3	6.44	1.15	1.07	16.68
G/S	7-98	49	290.55	17.05	34.60*	4-101	52	294.77	17.17	32.88*	7-104	55	294.7	17.17	31.09*
TGW	10-52	36	66.14	8.13	22.33*	14-56	40	66.14	8.13	20.12*	16-82	42	74.02	8.60	20.20*
SG	0-9	4.71	7.01	2.34	77.17*	0-9	4.81	8.01	2.89	67.14*	0-9	4.50	7.80	2.72	57.74*
VW/L	0-5	4.33	0.93	0.51	28.14	0-5	5	0.73	0.59	22.4	0-5	4.73	0.63	0.79	18.34

PH: (Plant Height), PE/L: (Peduncle Extrusion length), P/L: (Peduncle Length), NT/P: (Number of Tiller per Plant), SL: (Spike Length), SL/S: (Spikelet per Spike), FLL: (Flag Leaf Length), FLW: (Flag Leaf Width), LA: (Leaf Area), GFD: (Grain Filling Duration), DH: (Days to Heading), DM: (Days to Maturity), Y/P: (yield per Plant), B/Y: (Biological Yield), HI: (Harvesting Index), SSGW: (Single Spike Grain Weight), SW: (Single Spike Weight), G/S: (Grain per Spike), TGW; (Thousand Grain Weight), SG; (Stay Green), VWL: (Visual Wax on Leaf)

Statistical analysis

Principal component analysis (PCA) and correlation analysis is frequently used in data sciences (Brown-Guedira *et al*., 2000). PCA or factor analysis is a dimensionality reduction technique that helps to capture the essence of data pattern in larger datasets into few principal components (Leilah & Al-Khateeb, 2005; Khodadadi *et al*., 2011; Janmohammadi *et al*, 2014). XLSTAT from (Addinsoft, 2019) was used to perform PCA. XLSTAT software was also used to perform agglomerative hierarchical clustering (AHC) and to construct dendrogram of agro morphological data. Analysis of Variance (ANOVA) was implemented by using R software.

Results

Statistical analysis of phenotypic data: A descriptive statistical summary of variable, such as range, mean, standard deviation (n), variance, and coefficient of variation (%CV) of both years (2017-18 to 2018-19) and their respective control were presented in (Table 3). However CV≤ 20 is considered as acceptable range for field experiments. Value of coefficient of variation (CV) is important indicator to quickly estimate the % variability associated with a trait in a population. In this experiment PEL (CV = 37%, 24%, 24%), NT/P (CV = 36%, 32%, 32%), LA (CV = 22%, 24%, 25%), Y/P (CV = 32%, 33%, 29%), B/Y (CV = 40%, 30%, 28%), HI (CV = 52%, 36%, 33%), SSGW (CV = 42%, 38%, 36%), G/S (CV = 34%, 32%, 31%) and TGW (CV= 22%, 20%, 20%) showed higher coefficient of variation in both growing season in comparison to control conditions respectively (highlighted with asterisk in Table 3). However, PH, PE/L, NT/P, LA, DM, Y/P, SSGW, and SW showed a reduction in mean values of traits during both season in comparison to control (Table 3). Mean values of all traits were compared (Fig. 2). All traits were also presented in the form of boxplot (Supplementary Fig. F1) that represented the behavior of traits under normal and stress conditions. Post anthesis yield related traits showed a noticeable difference in mean in both growing season among treatment and control as evident from the results of ANOVA. Most of the traits were significantly different from control conditions results. All genotypes were evaluated and the genotypes in (Table 4A) showed a minimum and (Table 4B) showed maximum values of all trait during both seasons under stress conditions in comparison to control.

Correlation analysis: Pearson correlation matrix of all traits under induced heat stress environment for both year (2017-18 and 2018-19) and controlled conditions are presented in (see Supplementary Tables 2A, 2B and 2C at the end) respectively. Plant height, showed a positive correlation with peduncle length followed by spike length and NT/P during both years 2017-18; (r=0.49), (r=0.48), (r=0.28), 2018-19; (r=0.51), (r=0.35), (r=0.21) control; (r=0.55), (r=0.47), (r=0.28) respectively. Plant height also showed a positive but nonsignificant correlation with PEL and SL/S. Spike length showed a positive correlation with spikelet per spike (r=0.43), (r=0.45) and (r=0.47) in both season and control respectively. A strong positive correlation among FFL, FLW and LA was observed during both growing seasons 2017-18; (r=0.43), (r=0.88), (r=0.79), 2018-19; (r=0.49), (r=0.87), (r=0.83), and control; (r=0.43), (r=0.83), (r=0.73) respectively. Moreover DH showed a positive correlation with DM during both cropping seasons treatment (r=0.32), (r=0.57) and control (r=0.57) respectively. Days to maturity showed a strong positive correlation with GFD (r=0.80), (r=0.60) and (r=0.60) during both seasons and control. Yield per plant showed a positive correlation with B/Y, SSGW, SW and TGW during both years and control conditions. Biological yield showed a strong negative correlation with HI during both year and control, (r=-0.69), (r=-0.49) and (r=-0.53) respectively. A strong positive correlation was observed among all these traits SSGW, SW, G/S and TGW under stress and control conditions. Stay green also showed a positive correlation with days to heading under all treatment and control. However VWL wasn't correlated with any other traits in the population during both growing years. All traits showing a noticeable value of correlation either positive or negative are highlighted in with asterisk (*) in correlation (see Supplementary Table 2A, 2B and 2C at the end).

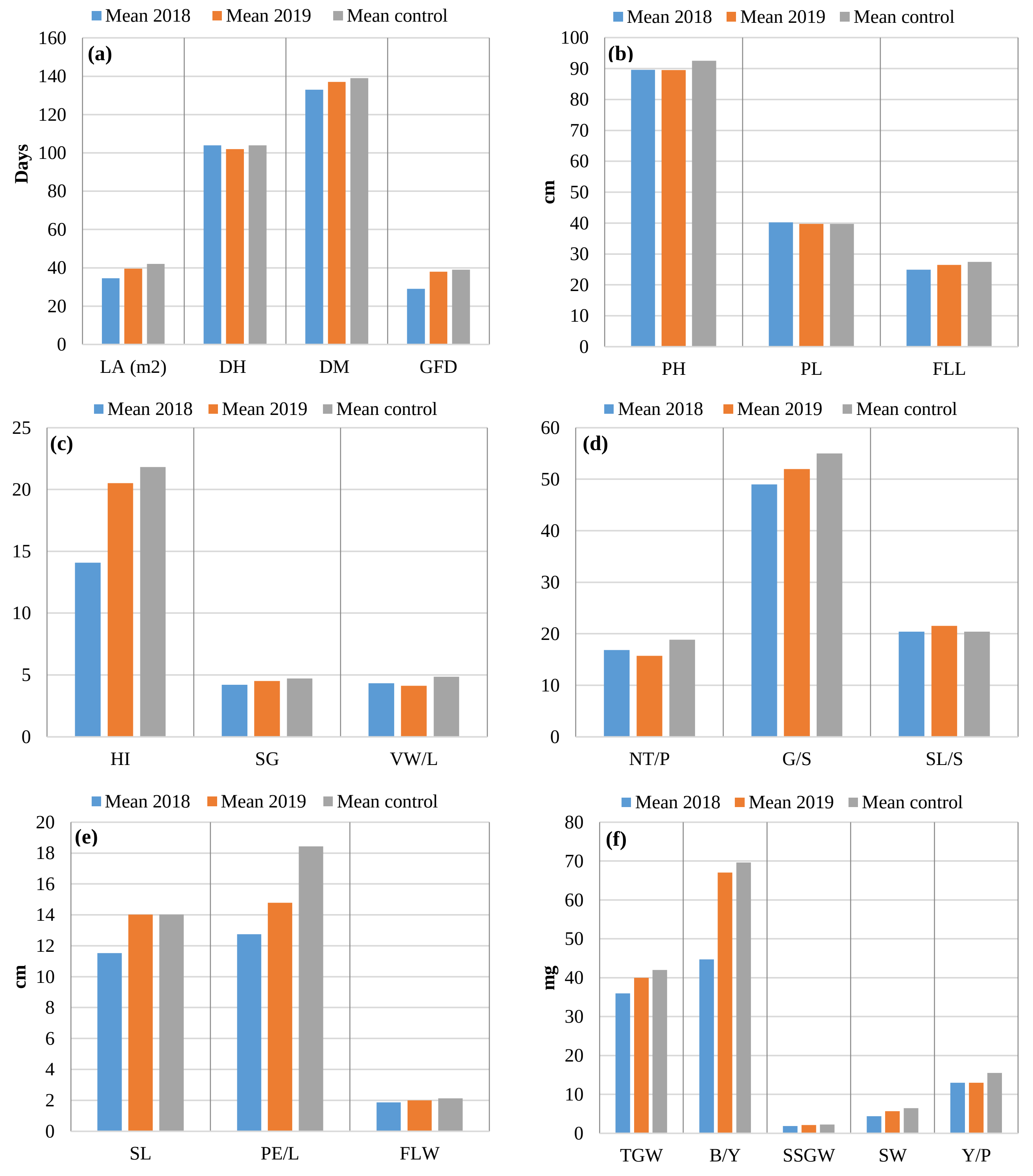
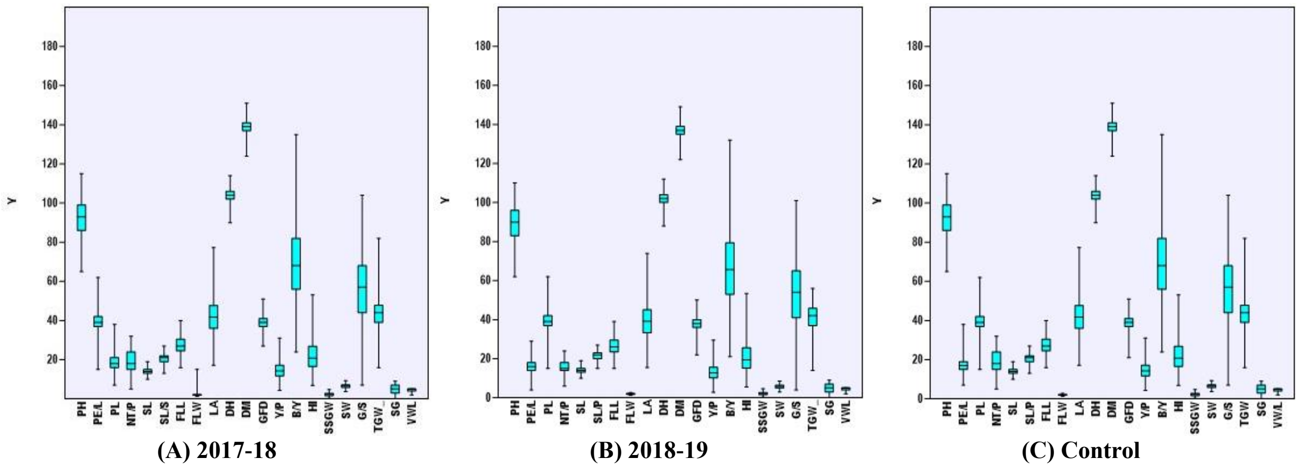


Fig. 2. Means of all traits in both year's treatment and control. Comparison of means of LA, DH, DM and GFD (a) Comparison of means of PH, PL and FLL (b) Indicating comparison of means of HI, SG and VW/L (c) Comparison of means of NT/P, G/S and SL/S (d) comparison of means of SL, PEL and FLW (e) Comparison of means of TGW, B/Y, SSGW, SW and Y/P during both years and control (f).



Supplementary Fig. 1. Box Plot of Agro-morphological traits of wheat crop under heat stress and control conditions.

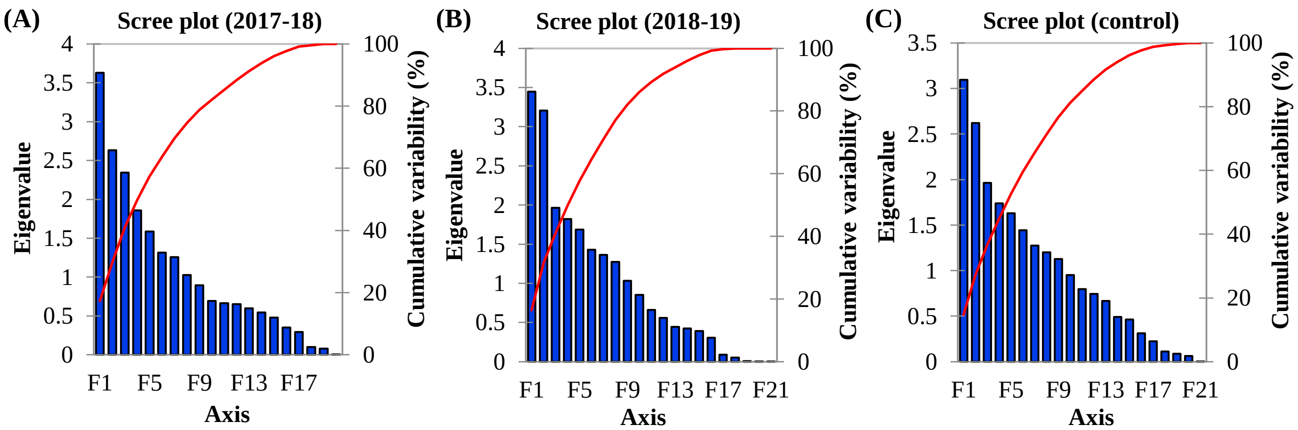


Fig. 3. Scree plot presenting principal factors on the base of eigenvalue and cumulative variability of agro-morphological and yield related traits, Scree plot of season 2017-18 (A) Scree plot 2018-19 (B) Showed scree plot of control (C).

Table 4. Genotypes showing minimum (A) and maximum (B) values of all traits during both growing season and control under stress conditions.

Traits		Genotype	2017-18	2018-19	Control	Traits		Genotype	2017-18	2018-19	Control
PH (cm)	Min	CB-512	62	62	65	DM (Day)	Min	CB-186	144	147	147
PEL (cm)	Min	CB-30	4	4	7	GFD (Day)	Min	CB-30	13	16	21
PL (cm)	Min	CB-422	12	15	15	Y/P (g)	Min	CB-186	3	3	4.5
NT/P (No)	Min	CB-512	3	11	5	B/Y (g)	Min	CB-198	18	27	30
SL (cm)	Min	CB-17	10	10	10	HI	Min	CB-328	5	6	7.9
SL/S (No)	Min	CB-17	13	15	13	SSWG (g)	Min	CB-249	1	0.4	0.5
FLL (cm)	Min	CB-332	15	15	16	SW (g)	Min	CB-328	1.7	3	3.7
FLW (cm)	Min	CB-332	1.5	1.4	1.4	G/S (No.)	Min	CB-349	7	4	11
LA (m^2)	Min	CB-332	20	16	17	TGW (g)	Min	CB-198	10	14	16
DH (Day)	Min	CB-392	90	88	90	VW/L	Min	CB-484	5	5	5

(A) Genotype showed a minimum value of all traits under stress conditions.

Traits		Genotype	2017-18	2018-19	Control	Traits		Genotype	2017-18	2018-19	Control
PH (cm)	Max	CB-320	112	110	115	DM (Day)	Max	CB-42	140	149	151
PEL (cm)	Max	CB-477	30	29	39	GFD (Day)	Max	CB-42	43	40	51
PL (cm)	Max	512	65	62	62	Y/P (g)	Max	CB-463	21	29	31
NT/P (No)	Max	CB-123	28	24	30	B/Y (g)	Max	CB-282	100	106	109
SL (cm)	Max	CB-268	18	19	19	HI	Max	CB-216	60	65	76
SL/S (No.)	Max	CB-373	27	25	27	SSGW (g)	Max	CB-427	4.3	4.6	4.7
FLL (cm)	Max	CB-429	38	39	40	SW (g)	Max	CB-23	7	8.5	9.3
FLW (cm)	Max	CB-208	2.2	2.6	2.6	G/S (No)	Max	CB-434	98	101	104
LA (m^2)	Max	CB-427	74	74	78	TGW (g)	Max	CB-11	52	56	58
DH (Day)	Max	CB-433	114	112	114	VW/L	Max	CB-69	2	2	2

(B) Genotypes showed a maximum value of all traits under stress conditions.

Table 5. Eigenvalue and cumulative variability of 205 Pakistani spring wheat genotypes for both cropping years and control season.

		F1	F2	F3	F4	F5	F6	F7	F8	F9
Eigenvalues	2017-18	3.629	2.633	2.342	1.860	1.587	1.313	1.257	1.025	1.011
Eigenvalues	2018-19	3.456	3.265	1.973	1.825	1.674	1.439	1.362	1.275	1.032
Eigenvalues	Control	3.436	3.205	1.963	1.821	1.684	1.429	1.362	1.275	1.032
Variability (%)	2017-18	17.282	12.539	11.153	8.856	7.558	6.250	5.988	4.882	4.801
Variability (%)	2018-19	16.411	15.261	9.349	8.670	8.020	6.803	6.486	6.073	4.913
Variability (%)	Control	14.741	12.487	9.348	8.278	7.769	6.875	6.063	5.714	5.366
Cumulative %	2017-18	17.282	29.822	40.974	49.830	57.388	63.638	69.626	74.508	76.501
Cumulative %	2018-19	16.411	31.673	41.021	49.692	57.712	64.515	71.001	77.074	81.987
Cumulative %	Control	14.741	27.228	36.576	44.854	52.623	59.498	65.561	71.275	76.641

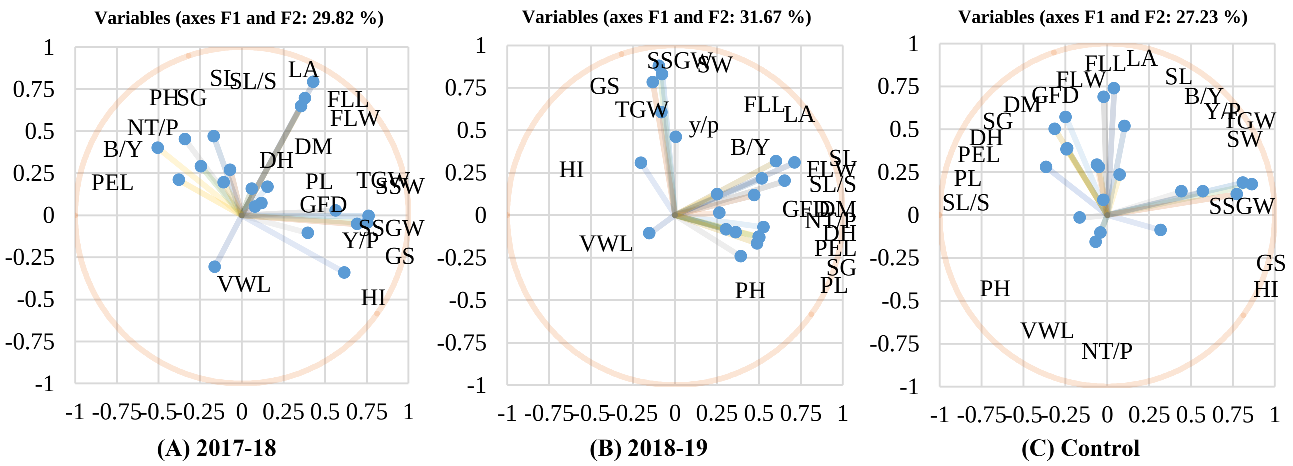


Fig. 4. Variable plot of both growing years and respective control.

Eigenvalue and cumulative variability (%): Eigen value of a principal Component (PC), a valuable tool to select the number of PCs is used for downstream data analysis. Hussain *et al.*, (2014) and Kaiser (1960) formulated a key criteria for the selection of PCs for further data analysis. In this experiment the principal component analysis (PCA) divided the variables or factors into 21 PCs in both growing seasons 2017-18, 2018-19 and their respective year control. Only PCs having eigenvalues ≥ 1 were selected for the bi-plot and other downstream analysis (Table 5).

Moreover out of 21 PCs, first 9 PCs explained 75% of cumulative variability associated with this data. During growing season 2017-18 first 9 PCs or factors having > 1 eigenvalues explained 76% cumulative variability in this season. While during growing season 2018-19 first 9 factors or PCs having >1 eigenvalue, and these 9 factors explained 82% cumulative variability during this cropping season. In case of control, mean of both years control, year one (2017-18) and year two (2018-19) control data was used to extract fruitful information and compare this information with the treatment data of both years. In case of control first 9 factors explained 77% cumulative variability associated with agro morphological and yield related traits. In nutshell first 9 worthwhile PCs were used for downstream bi-plot construction and data analysis.

Scree plot: It is a line plot that displays variables or factors on x-axes and eigenvalue of variables on y-axes that make an elbow shaped curve. The slope of the curve indicates the point or number of PCs to be used for construction of bi-plots. Three different scree plots of both growing seasons (2017-18) are presented in Fig. (3A), (2018-19) Fig. (3B) and control (Fig. (3C). It was clearly observed from the plots that first 9 PCs explaining >75% cumulative variability were used to draw bi-plot.

Bi-plot of variables: Correlation of 21 agro morphological variables were presented in a circular plot. Correlation among variables is depicted on the base of length of vector from the origin as well as on an angle between the variables. Acute angle < 90° depicts a positive correlation, whereas acute angle < 45 indicates a strong positive correlation among variables. On the other hand obtuse angle > 90 indicates negative correlation and obtuse angle >135° and <180° indicates a strong negative correlation among variables. However right angle (90°) variables showed no correlation. Length of a vector from its origin indicates the extent of variability explained by these variable as evident from (Fig. 4A, 4B and 4C) representing the variable plot of both years treatment and control season.

Bi-plot of season 2017-18: Bi-plot was divided into four groups on the bases of values of variables and factors loadings in both axes of a PCI and PC2. First group showed positive values of factor loadings and variables in both factors F1 and F2 in bi-plot. However, in 2nd group F1 showed negative value for PC1 and F2 showed positive value for PC2 of variables and factor loadings in both PCs. Third group showed positive value for F1 in PC1 as well as negative value of F2 in PC2 for variables and factor loadings. On the other hand fourth group showed a negative value for both PCI and PC2 variables and factor loadings in bi-plot. First and second group were collectively designated as positive groups of bi-plot and third and fourth groups were considered as negative coordinates of this bi-plot respectively. Vector length of variables determined the extent of variability associated with each variable. Shorter length variables are less divers compared to larger, that grasps more diversity. All observations that associated with a variable are also dispersed in similar direction of subjected variable in a bi-plot.

In this season (2017-18) first two axes F1 and F2 explained 29.82% variability associated with these variables. Both F1 and F2 explained 17.3% and 12.5% variability respectively which were used to construct bi-plot of variables. In (Fig. 5.) PH, SL, BY, PEL, FFL, FLW, LA, SW, SSGW, G/S, TGW, Y/P and HI held a larger vector length which was directly associated with larger amount of variability explained by these variables. However DH, DM, SL/S, GFD, NT/P VWL, SG and PL having shorter vector length which was directly associated with small amount of variability associated with these traits. All 205 genotypes were divided into four groups of bi-plot. During cropping season 2017-18, (44), (50), (49), (58) genotypes were divided in (group 1), (group 2), (group 3) and (group 4) of this bi-plot respectively as showed in (Fig. 5).

Bi-plot of season 2018-19: In cropping season 2018-19 about 31.67% variability of different variables were explained with F1 and F2 axes in Figure 6. Both F1 and F2 explained 16.41% and 15.26% of total variability respectively, which were used for the construction of bi-plots of these variables. On contrary to the last year results PH, SL, BY, PEL, FFL, FLW, LA, SW, SSGW, G/S TGW, Y/P, SG and NT/P having larger vector length which were directly associated with larger amount of variability linked to these variables. Moreover DH, DM, GFD, VWL, HI, SG and PL showed smaller vector length which meant lesser variability explained by these variables. In cropping season 2018-19 (54), (58), (42), (55) genotypes were divided into four groups (group 1), (group 2), (group 3) and (group 4) of bi-plot respectively (Fig. 6).

Bi-pot of control 2017-18 and 2018-2019: For control analysis, mean data of both years control was used to construct bi-plot. First two axes of the bi-plot explained 27.23% variability in agro-morphological traits of this control population. Both F1 and F2 explained 14.74% and 12.49% variability associated with these traits. In control data G/S, SSGW, SW, TGW, Y/P, SL, LA, FLL, DM, PEL, PL and HI having larger vector length which meant greater variability associated with these variables. However, PH, VWL, NT/P, B/Y, GFD, FLW and SL/S having smaller vector length, meaning lesser variability associated with these traits. In case of control season (49), (57), (44), (59) genotypes were classified into four separate groups (group 1), (group 2), (group 3) and (group 4) respectively as depicted from (Fig. 7.). Moreover 14 genotypes CB-42, CB-94, CB-176, CB-200, CB-208, CB-210, CB-239, CB-247, CB-350, CB-373, CB-399, CB-420, CB-484 and CB-498 showed a consistent behavior and positive correlation with yield and other variables of this group during both cropping season and control. On the other hand group 4 of bi-plot contained 58, 55 and 59 genotypes in both growing seasons and control respectively. However 13 genotypes of group 4 such as CB-11, CB-18, CB-60, CB-124, CB198, CB-242, CB-248, CB-332, CB-372, CB-381, CB-443, CB-466 and CB-522 showed a negative correlation with yield and other variables of group 1 of the bi-plot during both growing seasons and control.

Table 6. Combined analysis of variance (ANOVA) for both years (2017-18 and 2018-19) of 21 agro-morphological traits of 205 Pakistani spring wheat genotypes.

SOV	Treatment	Year	Genotype	Rep	Treat: Year	Treat : Genot	Year: Genot	Rep: Block	Residuals
DF	2	1	204	2	1	204	204	3	1848
PH	111227331***	22559***	816***	526***	36683***	137***	137	360	16
PE/L	805106***	122^{ns}	148***	3401***	485**	41^{ns}	19^{ns}	65^{ns}	70
P/L	1330009***	1881***	257***	6573***	4^{ns}	22^{ns}	4^{ns}	204*	65
NT/P	271911***	368***	59***	3704***	158***	12^{ns}	13^{ns}	75***	12
S/L	277165***	41^{ns}	84***	2246***	226***	7^{ns}	35	77***	14
SL/S	513749***	90***	38***	0	90***	6	6	1^{ns}	1
FLL	867262***	0	138***	286***	1764***	22***	22	2^{ns}	4
FLW	4898.5***	1.3***	3***	0.3***	2.4***	2.8***	0.1	0	0
LA	1941575***	909**	1091***	295*	6892***	478***	141	15^{ns}	89
DH	12913275***	70**	414***	312***	1098***	71***	18	11^{ns}	7
DM	2306385***	5698***	176***	51***	2569***	30***	30	51^{ns}	6
GFD	1555332***	10492***	401***	632***	8867***	54***	54	15^{ns}	7
Y/P	245034***	497***	218***	261^{ns}	3^{ns}	0	0	175***	1
B/Y	4514380***	91366***	2974***	2593***	72470***	542***	519	3^{ns}	69
HI	848209***	37214***	1030***	2795***	27581***	350***	328	450^{ns}	47
SSGW	6004.5***	50.7***	7.8***	5.3***	12.7***	0	0	0.1***	0
SW	41558***	263***	14***	12***	162***	0	0	3***	0
GS	3538189***	10429***	3096***	4772***	12185***	24^{ns}	24^{ns}	70^{ns}	43
TGW	1828123***	24782***	696***	15416***	31715***	30^{ns}	6^{ns}	118^{ns}	73
SG	26895***	2.4^{ns}	74.7***	2.4^{ns}	2.4^{ns}	0.7^{ns}	0.7^{ns}	0.9^{ns}	0.8
VW/L	21156.1***	0	6.9***	0	0	0	0	5.5***	0

Significance codes: '***'p< 0.001 '**'p< 0.01 '*'P< 0.05, ns= nonsignificant.

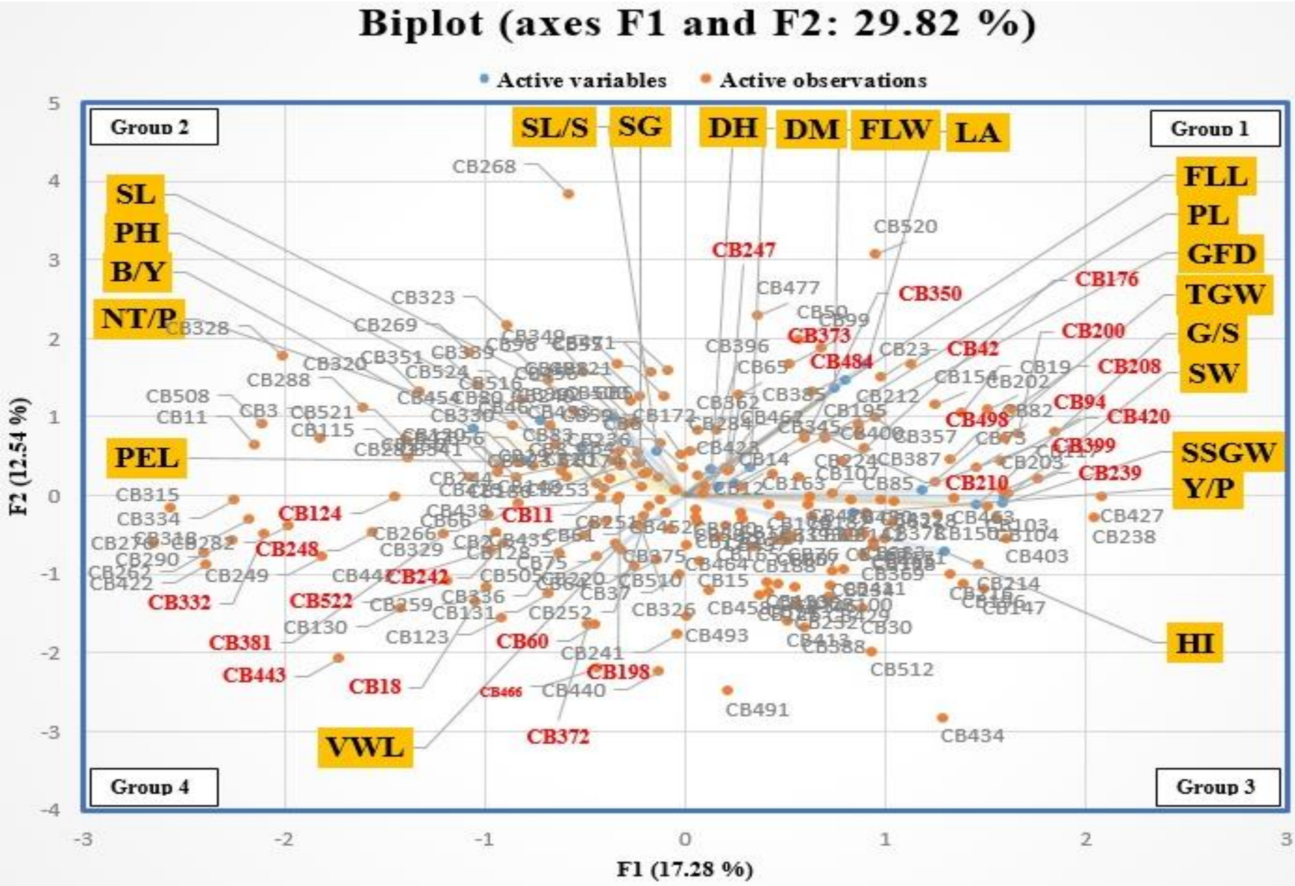


Fig. 5. Bi-plot of variables and observations growing season 2017-18.

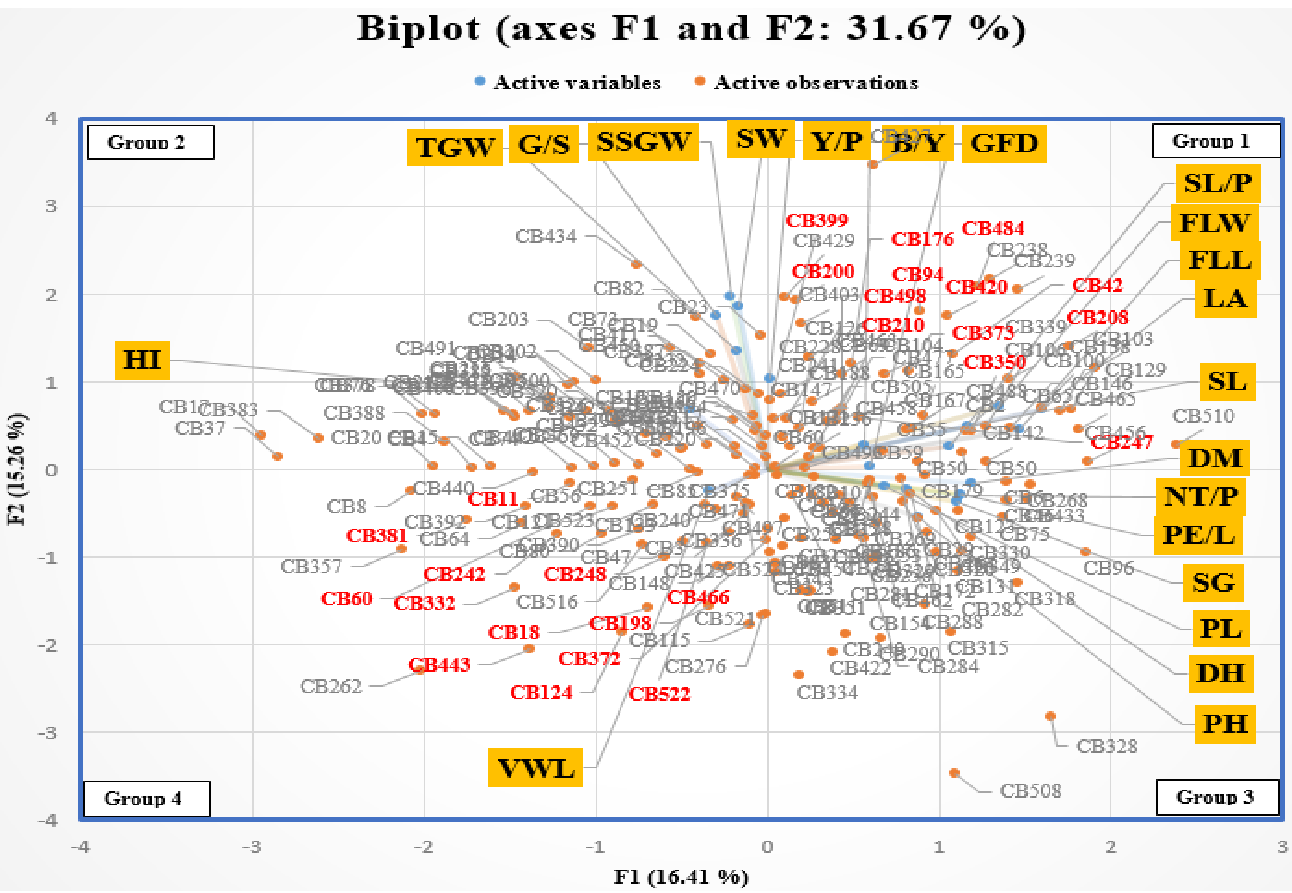


Fig. 6. Bi-plot of variables and observations of growing season 2018-19.

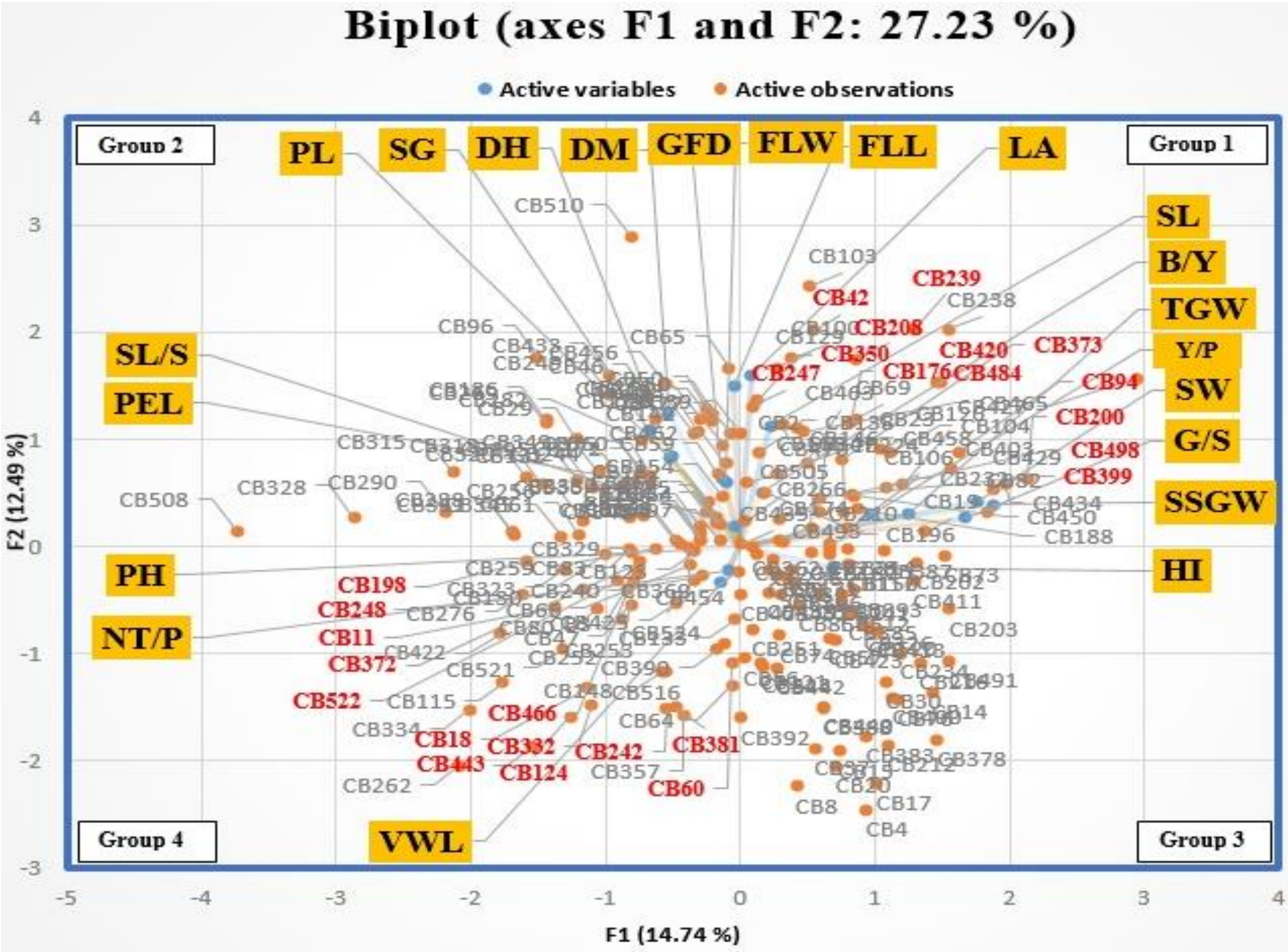


Fig. 7. Bi-plot of variables and observations of control season.

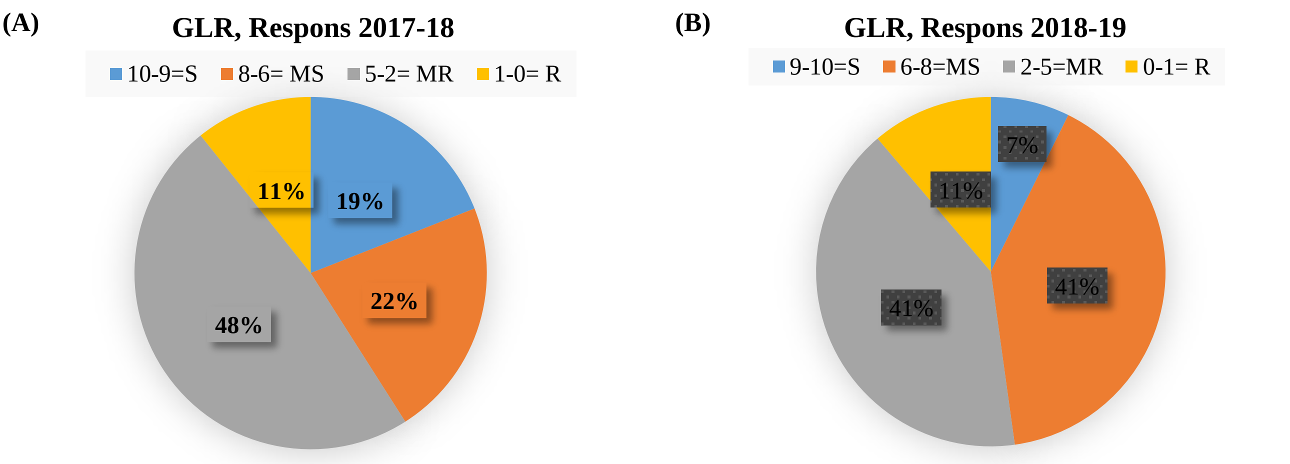


Fig. 8. Response of genotype on GLR scale, (A) Year 2017-18, (B) Year 2018-19.

Analysis of variance (ANOVA): Combined analysis of variance (ANOVA) of both years treatment and control was presented (Table 6). Combined ANOVA was performed on 6 replication per year (3 rep of treatment and three rep of control) and experiment was repeated for two consecutive years 2017-18 and 2018-19. Most of the agro morphological traits were depicted that induced heat stress possessed a significant influence on phenotypic expression of all traits. Most of the traits were significant at $p<0.001$.

Green leaf retention (GLR): Stay green or green leaf retention (GLR) is an important method used to access the ability of a genotype to retain green leaf area after brief period of heat stress (Borrell *et al*., 2000; Joshi *et al*., 2007). The ability to stay green is directly linked with the rate of photosynthesis and movement of phyto-assimilates from source to sink in plant. Only a single observation was recorded during each year after removal of induced heat stress conditions. On the base of GLR scale, all genotypes (205) were divided into 4 groups during year 2017-18 containing 22

(11%), 99 (48%), 45 (22%) and 39 (19%), while during 2nd year 2018-19 containing 23 (23%), 84 (41%), 83 (41%) and 15 (7%), genotypes were designated as resistant (R), moderately resistant (MR), moderately susceptible (MS), and susceptible respectively (Fig. 8.). Response of different genotypes during both years and control season given in (see Supplementary Table 3 at the end).

Heat susceptibility index (HSI): Heat susceptibility index is another imperative method to screen germplasm against terminal heat stress in wheat and other field crops (Rahman *et al*., 2009; Oliveira *et al*., 2011). According to the HSI, whole population was divided into three groups. Out of 205, 8 genotypes CB-11, CB-19, CB-103, CB-216, CB-228, CB-373, CB-400 and CB-403 showed HSI value of (0.23, 0.34), (0.24, 0.34), (0.30, 0.37), (0.34, 0.25), (0.21, 0.32), (0.20, 0.17), (0.42, 0.37) and (0.45, 0.31) respectively for both growing seasons 2017-18 and 2018-19. Genotypes showed HSI≤ 0.5 that was considered as high heat tolerant. Heat tolerant genotypes were highlighted with asterisk (*) in the (see Supplementary Table 4 at the end). However 87 genotypes showed a HSI value ranges between 0.5-1.0 which were considered as moderately tolerant. Total of 110 genotypes having HSI≥ 1 were considered as heat susceptible as mentioned below in (see Supplementary Table 4 at the end). Heat susceptibility index of both years 2017-18 and 2018-19 indicated a quite similar response of genotypes during both season (Fig. 9).

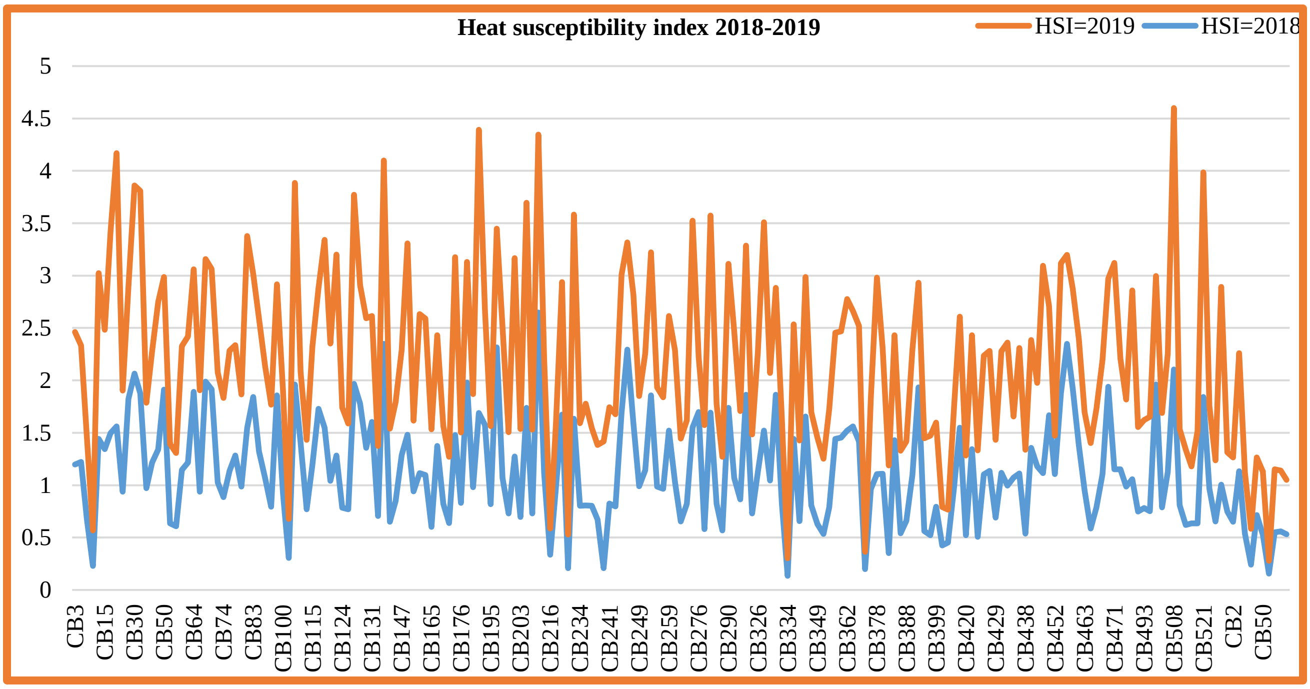


Fig. 9. Heat susceptibility index value of year 2017-18 and 2018-19.

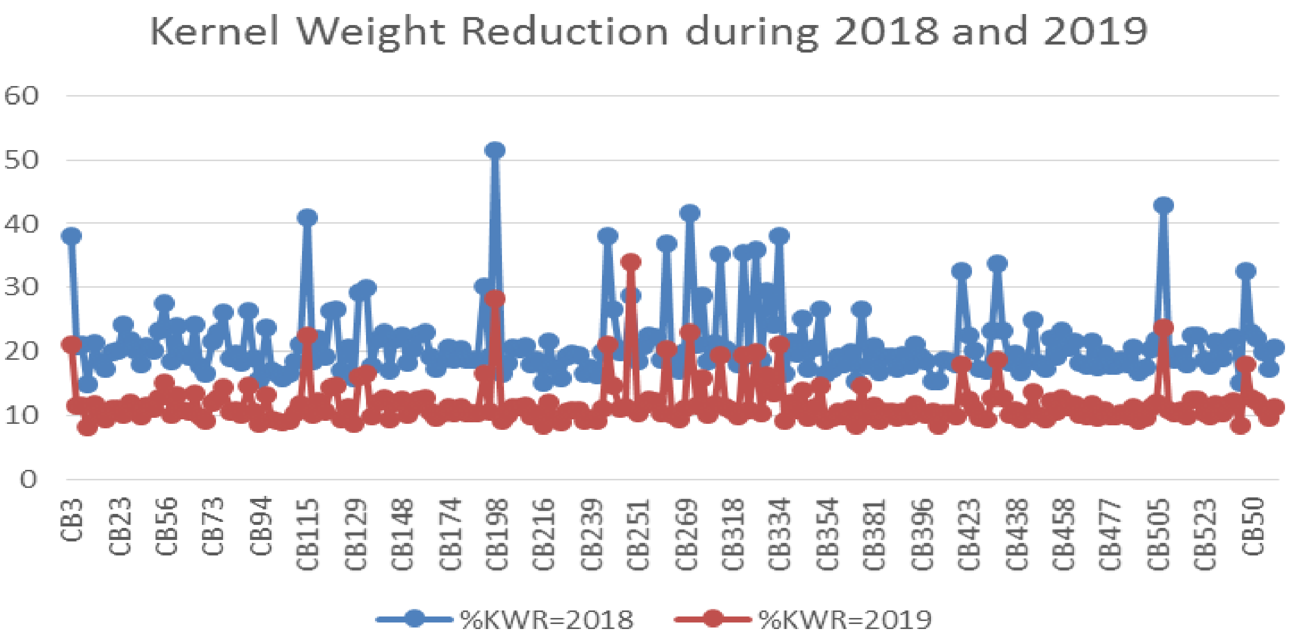


Fig. 10. Kernel weight reduction of year 2017-18 and 2018-19.

Kernel weight reduction (KWR): All genotypes showed a marked reduction in TGW during both growing seasons 2017-18 and 2018-19. Out of 205, merely 8 genotypes i.e. CB-11, CB-19, CB-103, CB-216, CB-228, CB-373, CB-400 and CB-403 showed ≤ 20% cumulative reduction in thousand grain weight (TGW) during both seasons (Table 10 highlighted with single asterisk (*)). These genotypes may be a potential source for stress breeding program in future. Selected lines may also prove as source of resistant genes for other functional genomic and fine mapping studies. Moreover about 179 genotypes showed < 50% but > 20% cumulative reduction in both seasons. However, 18 genotypes, i.e. CB-3, CB-18, CB-115, CB-198, CB-242, CB-249, CB-262, CB-276, CB-290, CB-232, CB-334, CB-422, CB-433 and CB-508 showed ≥ 50% cumulative reduction in TGW during both cropping seasons 2017-18 and 2018-19 (see Supplementary Table 5 at the end with two asterisk **). KWR index of both seasons showed a consistent behavior of all genotypes during both years (Fig. 10).

Agglomerative hierarchical clustering (AHC) of agro-morphological data: Euclidean distance based Ward's method was used for the cluster analysis of 205 local genotypes. The whole population was divided into three clusters C1, C2 and C3 during each cropping season 2017-18, 2018-19 as well as in respective control. However, in cropping season 2017-18, all genotypes were divided into cluster C1, C2 and C3 containing 75, 63, 67 genotypes respectively (Fig. 11). On the other hand in 2nd cropping season the whole population was also divided into three clusters having 52, 91, 62 genotypes in cluster C1, C2, and C3 respectively (Fig. 12). On contrary to both cropping seasons in control the whole population was also divided into three cluster C1, C2, C3 having 41, 79, 85 genotypes in each cluster (Fig. 13). This type of clustering will be helpful for parent's selection for future breeding programs and to infer the similarity index of genotypes in a population.

Discussion

Weather fluctuation directly associated with crop development and ultimately leads toward reduction in quality as well as final genetic gains of crops, which is continuously threatening global food security (Martiniello and Teixeira de silva, 2011; Hakim *et al*., 2012; Hossain *et al*., 2012; Hossain and teixeira da silva, 2012). This experiment was designed to estimate genetic diversity in agro-morphological traits as well as to harness the response of induced terminal heat stress on yield parameters in Pakistani genotypes. It was observed that sudden heat shock in timely sown and expected systematic heat wave to late sown crop imposed a significant negative impact on morphological as well as on yield contributing traits at terminal growth stage. Most of the yield associated post-anthesis traits i.e. TGW, G/S, SW, Y/P, SSGW, B/Y, HI, GFD, VWL, SG, LA and DM showed a significant reduction under heat stress condition compared to normal conditions. However, morphological traits such as PH, PL, PEL, NT/P, DH, SL and SL/S did not show any significant change under induced terminal heat stress environment.

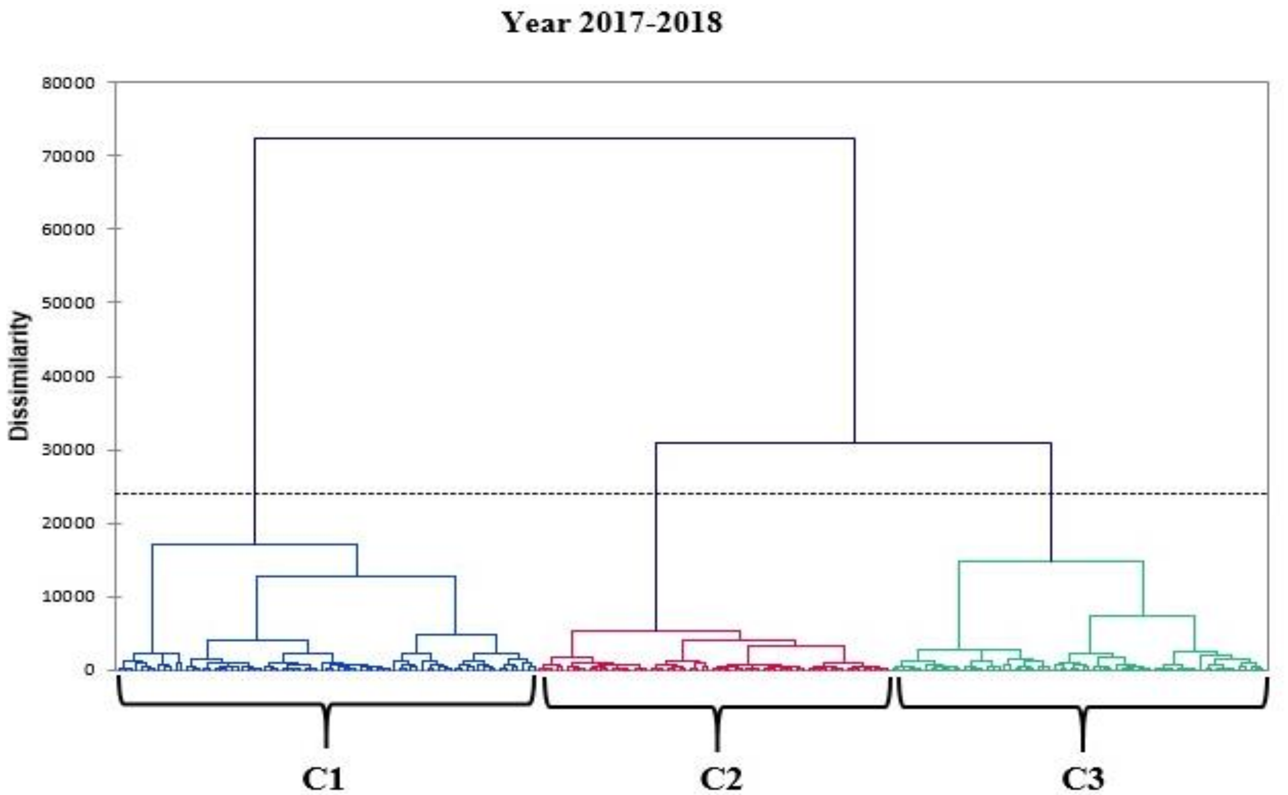
Descriptive statistical values of range, minimum, maximum, mean, variance, standard deviation (n) and coefficient of variation (%CV) of 21 agro morphological traits provided a swift bird eye view of the population data. In current era descriptive statistics become a norm for the dissection of quantitative morphological and yield related traits (Din *et al*. 2018; Gulnaz *et al*., 2019; Iqbal *et al*., 2017; Pooja *et al*., 2018; Shahid *et al*., 2017; Yaqoob, 2016). It was clear from the range of the data that a huge amount of variability in all morphological and yield related traits were present. This variability revealed the presence of ample amount of potential to improve these traits with respect to various biotic and abiotic stresses. Both, control and treatment were raised under same crop husbandry before the application of heat treatment. It is evident from the descriptive statistics that post-anthesis traits showed a considerably higher CV in comparison to control in both cropping season 2017-18 and 2018-19.

In this study plant height have a positive correlation with, peduncle length (Habib *et al*., 2020) peduncle extrusion length, number of tiller per plant and spike length. Spike length have a positive correlation with spikelet/spike. DH showed a positive correlation with DM, but negative correlation with GFD. Days to maturity showed a strong positive correlation with grain filling duration (Gulnaz *et al*., 2019; Ibrahim *et al*., 2008; Nawaz *et al*., 2013; Shahid *et al*., 2017). Flag leaf acts as a key organ responsible for synthesis and transport of phyto-assimilates from source to sink at all growth stages of plant. It ultimately leads towards growth as well as final genetic gains (Yield) in cereal crops. Agro-morphological attributes of flag leaf, such as FLL, FFW and LA are important factors to harness stress signals of adaptation and selection of plants holding desired traits. The results of current experiment FLL, and FLW, showed a strong positive correlation with LA (Biswal and Kohli, 2013; Fan *et al*., 2015; Tian *et al*., 2014; Tsukaya, 2006).

Moreover in current experiment yield per plant had a positive correlation with thousand grain weight, biological yield, harvesting index, spike weight, and single spike grain weight (Akram *et al*., 2008; Dogan, 2009; Ghaderi *et al*., 2009; Ibrahim *et al*., 2008; Kandic *et al*., 2009; Kashif & Khaliq, 2004; Khan *et al*., 2015; Subhashchandra *et al*., 2009). However, biological yield showed a negative correlation with harvesting index and spike weight. Grain per spike showed a significant positive correlation with single spike grain weight, (Mi *et al*., 2000; De Vita *et al*., 2007; Zafarnader *et al*., 2013; Djuric *et al*., 2018). Single spike grain weight showed a strong positive correlation with G/S and thousand grain weight. Grains per spike showed a significant positive correlation with thousand grain weight (Djuric *et al*., 2018; Jocković *et al*., 2014; Leilah & Al-Khateeb, 2005).

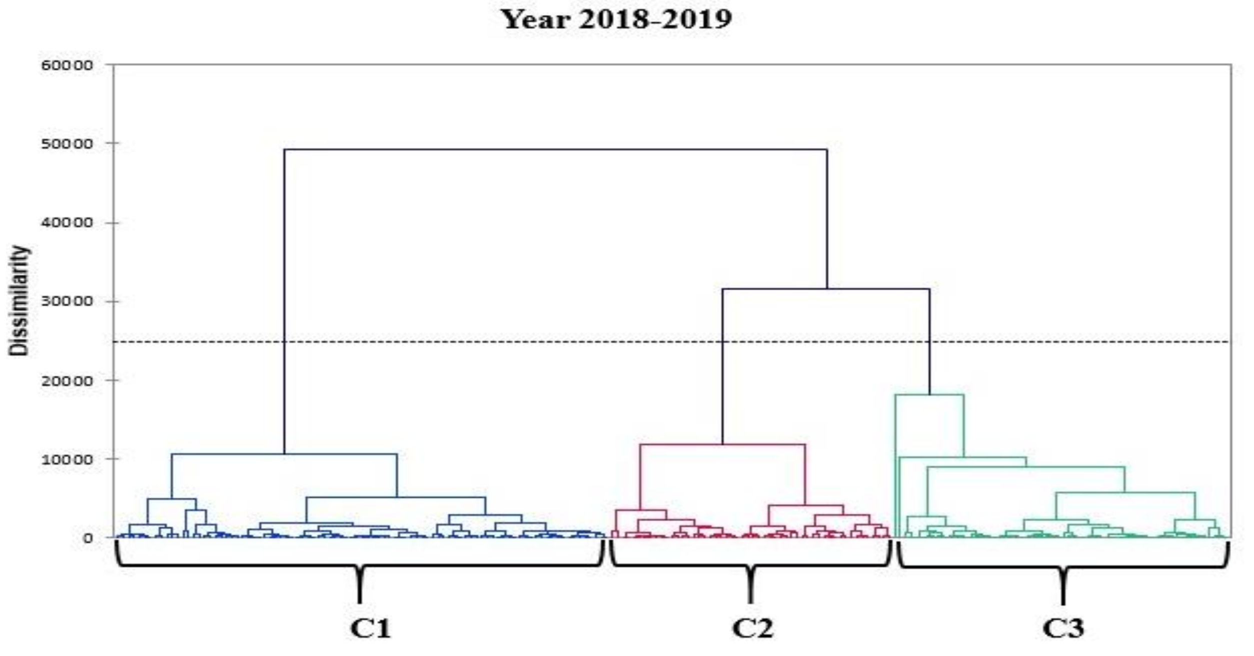
Green leaf retention (GLR) means the ability of a plant to retain green color for a longer period of time under stress condition especially in case of heat stress (Joshi *et al*., 2007). GLR is directly linked with photosynthesis and grain development from anthesis to late maturity during whole grain filling duration in stress conditions (Thomas and Howarth, 2000). It's a fast track method to score genotypes for GLR in field (Wanous *et al*., 1991). In current experiment most of the genotypes did not show consistent behavior for GLR score during both growing season. GLR showed a positive correlation (r=0.32) with DM in this population, but GLR was not correlated with plant yield under heat stress conditions (Joshi *et al*., 2007; Li *et al*., 2018). In our study GLR was not associated with heat stress tolerance, because both growing season all genotypes showed a variable response. Some genotypes retained the green colour for a longer period of time, but these genotypes did not provide any advantage to green plants over less green genotypes. However while studying GLR phenotype, it was observed that genotypes retaining green colour for longer period of time might help the plant secondary tiller to become fertile and try to compensate the yield losses at some extant after removing the heat stress conditions. However, this hypothesis needs further studies to confirm this notion.

Heat susceptibility index (HSI) is a quick method to evaluate the performance of wheat genotypes under stress and control conditions, it does not merely consider the yield under stress conditions (Kaur *et al*., 2009). HSI directly represent the ability of a genotype to escape and maintain its performance in case of stress by accelerating various morphological and cellular metabolic processes. The results of current experiment showed that, 8 genotypes CB-11, CB-19, CB-103, CB-216, CB-228, CB-373, CB-400 and CB-403 showed HSI value of 0.23-0.34, 0.24-0.34, 0.30-0.37, 0.34-0.25, 0.21-0.32, 0.20-0.17, 0.42-0.37 and 0.45-0.31 respectively for both growing seasons 2017-18 and 2018-19 these genotypes could be a potential source for gene discovery, functional genomic studies and various other crop breeding strategies. Sustainability in genetic gains under late sown or stress conditions conform the potential of genotypes to produce higher yield under elevated temperature regimes. On the other hand these varieties also showed a batter adaptability, more number of tiller per plant, higher photosynthetic rate, more grain per spike and grain weight, similarly different researcher (Ahmad *et al*., 2003; Villegas *et al*., 2007; Mason *et al*., 2010; Khan *et al*., 2015) also obtained similar kind of results in their studies.



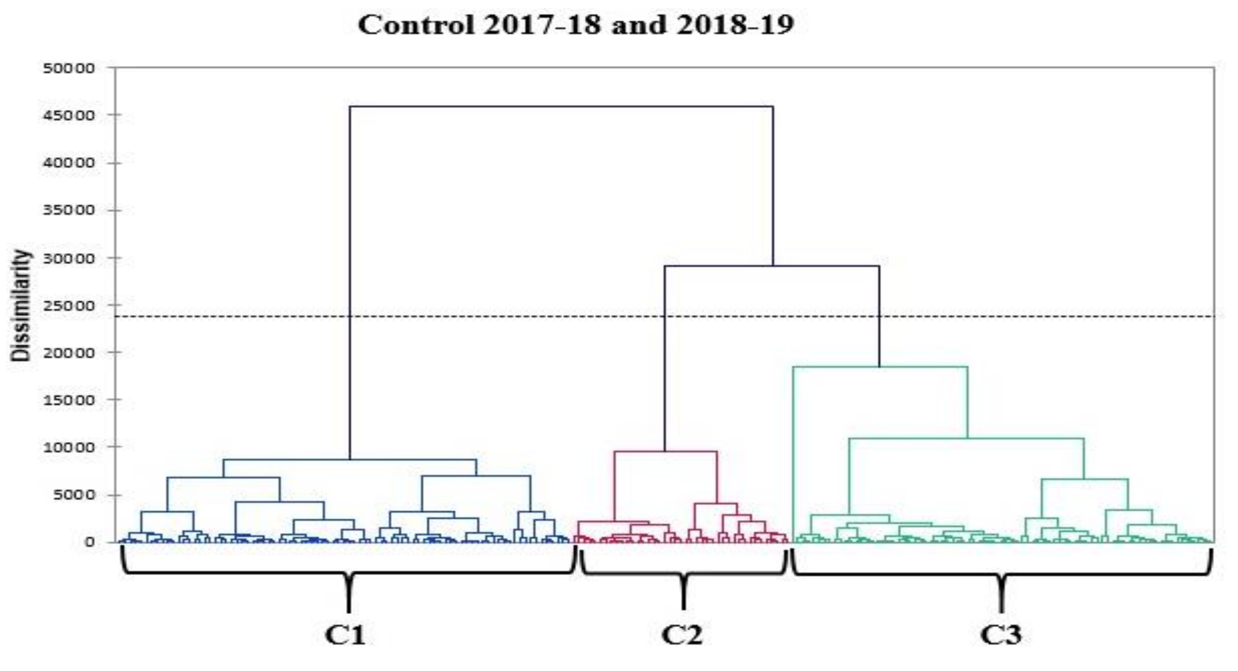
Class	Objects	Sum of weights	Within Class Variance	Minimum Distance to Centroid	Average Distance to Centroid	Maximum Distance to Centroid
1	75	75	1120.930	16.069	31.830	55.484
2	63	63	552.707	11.173	22.843	37.950
3	67	67	953.367	16.256	29.666	58.331

Fig. 11. Cluster analysis of 205 Pakistani wheat genotypes for year 2017-18.



Class	Objects	Sum of weights	Within-Class Variance	Minimum Distance to Centroid	Average Distance to Centroid	Maximum Distance to Centroid
1	52	52	944.196	11.306	29.178	53.337
2	91	91	701.631	12.329	24.888	60.920
3	62	62	1152.926	11.945	28.169	133.442

Fig. 12. Cluster analysis of 205 Pakistani wheat genotypes for year 2018-19.



Class	Objects	Sum of weights	Within-Class Variance	Minimum Distance to Centroid	Average Distance to Centroid	Maximum Distance to Centroid
1	41	41	954.454	15.278	29.351	52.581
2	79	79	968.315	11.794	27.098	135.359
3	85	85	869.798	13.818	27.870	58.982

Fig. 13. Cluster analysis of 205 Pakistani wheat genotypes for year 2017-18 (Control).

Conclusion

Wheat is one of the leading food crop for 2.5 billion peoples of 89 countries. Promptly increasing global population possessing a huge challenge in front of policy maker and plant scientists to ensure world food security under this era of swift climate change. Conclusively there is need to speed up the process of wheat germplasm evaluation to fast track the wheat breeding to develop climate resilient crops and final genetic gains. All post-anthesis traits were adversely effected by induced terminal heat stress such as HI, BY, Y/P, SW, G/S, TGW, SSGW, GFD, DM etc. Heat susceptibility index (HSI) as well as Kernel weight reduction (KWR) are two important tools for the screening of wheat germplasm. Some total of 8 genotypes CB-11, CB-19, CB-103, CB-216, CB-228, CB-373, CB-400 and CB-403 showed good response under stress condition and these genotypes could be a good source for the development of heat tolerant verities for local heat affected areas. However, green leaf retention (GLR) is a dynamic trait and showed a huge variability in both years (2017-18 and 2018-19). In a nutshell an ample amount of agro-morphological traits diversity exists in Pakistani local germplasm. In spite of such immense diversity, there is a room for further improvement of local germplasm against various biotic and abiotic stress by exploiting the findings of current experiment. Now a day’s genetic dissection of germplasm is a valuable tools for the identification of genes/QTLs associated with disease resistance or stress tolerance related traits. This phenotypic data of local lines will be further used for association mapping studies by using advanced genome wide association studies (GWAS) tools. These successfully mapped genes/ QTLs will be further finely mapped and tried to incorporate these findings for the development of climate resilient advanced breeder lines to ensure national as well as global food security.

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Supplementary Table 1. Supplementary data of serial number and pedigree of all genotypes with their respective CB # and origin.

Sr. No.	PEDGREE Local Lines (205)	CB #	Origin
1.	BHAKKAR-2000	CB3	Pakistan
2.	CHAKWAL-50	CB4	Pakistan
3.	FAREED-06	CB8	Pakistan
4.	INQILAB 91	CB11	Pakistan
5.	MANTHAR	CB14	Pakistan
6.	MIRAJ-08	CB15	Pakistan
7.	SHAFAQ-06	CB17	Pakistan
8.	BARS-09	CB20	Pakistan
9.	BHITTAI	CB23	Pakistan
10.	SASSI	CB30	Pakistan
11.	PIRSABAK 2005	CB37	Pakistan
12.	CHENAB-79	CB42	Pakistan
13.	IQBAL2000	CB46	Pakistan
14.	JAUHAR-78	CB47	Pakistan
15.	KOHISTAN 97	CB50	Pakistan
16.	PASINA 90	CB56	Pakistan
17.	PUNJAB-76	CB57	Pakistan
18.	PUNJAB 96	CB60	Pakistan
19.	SA-42	CB61	Pakistan
20.	SHAHKAR 95	CB65	Pakistan
21.	SHALIMAR-88	CB66	Pakistan
22.	ZA-77	CB69	Pakistan
23.	KAUZ'S'	CB73	Australia CIMMYT
24.	NACOZARI F-76	CB74	CIMMYT
25.	OASIS F-86	CB75	CIMMYT
26.	PBW-343=ATTILA	CB76	India=CIMMYT
27.	FRET-1	CB80	CIMMYT
28.	WH-542	CB82	India CIMMYT
29.	HOOSAM-3	CB83	ICARDA
30.	SAAR	CB85	CIMMYT
31.	CHAM-4	CB94	ICARDA
32.	CHILERO=CHIL'S'	CB96	CIMMYT
33.	FRONTANA	CB99	Brazil CIMMYT
34.	HARTOG=HTG.(PAVON)	CB100	CIMMYT
35.	OASIS/SKAUZ//4*BCN/3/2*PASTOR	CB103	CIMMYT
36.	BABAX/LR42//BABAX*2/3/VIVITSI	CB104	CIMMYT
37.	PBW 343*2/KUKUNA	CB106	CIMMYT
38.	PBW 343*2/KURUKU	CB107	CIMMYT
39.	PVN//CAR422/ANA/3/KAUZ*2/TRAP//KAUZ	CB115	CIMMYT
40.	TRAP#1/PBW65/3/KAUZ*2/TRAP//KAUZ	CB117	CIMMYT
41.	PVN/PBW65/3/KAUZ*2/TRAP//KAUZ	CB120	CIMMYT
42.	PARULA=PRL	CB121	CIMMYT
43.	NING-8319	CB123	China CIMMYT
44.	HARRIER 17.B	CB124	Australia CIMMYT
45.	V-03007	CB126	Pakistan
46.	CON.'S'/ANA 75//CON.'S'	CB128	CIMMYT
47.	HD2236//SA.42/HARRIER'S= V-97088	CB129	Pakistan
48.	PB81//F3.71/TRM/3/BULBUL// F3. 71/ TRM =V0005	CB130	CIMMYT
49.	WEEBILL-1 = V-03158	CB131	CIMMYT
50.	WATAN/2*ERA	CB133	CIMMYT
51.	TURACO/PRINIA	CB138	CIMMYT
52.	PB-96/87094//MH-97	CB146	Pakistan

Supplementary Table 1. (Cont'd.).

Sr. No.	PEDGREE Local Lines (205)	CB #	Origin
53.	MAYA/PVN	CB147	CIMMYT
54.	WL 711/CROW "S"//ALD #1 / CMH77A.917/3/HI 666/PVN 'S'	CB148	CIMMYT
55.	PRL/2*PASTOR//PARUS/5/NAC/TH.AC//3*PVN/3/MIRLO/BUC/4/2*PASTOR	CB150	CIMMYT
56.	FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ/5/ONIX	CB154	CIMMYT
57.	CROC-1/AE.SQ(224)//OPATA/3/FLAG-7	CB163	CIMMYT
58.	SERI.1B*2/3/KAUZ*2/BOW//K	CB165	CIMMYT
59.	SHUHA-4//NS732/HER/3/ MILAN/DUCULA	CB167	CIMMYT
60.	BWP 122526	CB171	Pakistan
61.	NW S-2001	CB174	Pakistan
62.	PR-111	CB176	Pakistan
63.	PR-106	CB179	Pakistan
64.	13248	CB186	Pakistan
65.	Long grain	CB188	Pakistan
66.	NSW-14	CB195	Australia CIMMYT
67.	V-12266	CB196	Pakistan
68.	V-13270	CB198	Pakistan
69.	122557	CB200	Pakistan
70.	V-02192	CB202	Pakistan
71.	V-02156	CB203	Pakistan
72.	V-04048	CB208	Pakistan
73.	V-05115	CB210	Pakistan
74.	V-06129	CB212	Pakistan
75.	V-056132	CB214	Pakistan
76.	V-06018	CB220	Pakistan
77.	V-06068	CB224	Pakistan
78.	KIRITATI//PBW65/2*SERI.1B	CB228	CIMMYT
79.	KIRITATI/4/2*SERI.1B*2/3/KAUZ*2/BOW//KAUZ	CB232	CIMMYT
80.	WHEAR/VIVITSI//WHEAR	CB234	CIMMYT
81.	WHEAR/CHAPIO//WHEAR	CB236	CIMMYT
82.	WHEAR/KUKUNA/3/C80.1/3*BATAVIA//2*WBLL1	CB238	CIMMYT
83.	INQALAB91*2/KUKUNA//	CB239	CIMMYT
84.	SUNCO//TNMU/TUI	CB240	CIMMYT
85.	SHARP/3/PRL/SARA//TSI/VEE#5/5/VEE/LIRA//BOW/3/BCN/4/KAUZ	CB242	CIMMYT
86.	DOLLARBIRD	CB244	Australia CIMMYT
87.	KIRITATI	CB247	CIMMYT
88.	PFAU/WEAVER*2//KIRITATI	CB248	CIMMYT
89.	PGO/SERI//BAV92	CB249	CIMMYT
90.	KINGBIRD#1	CB251	CIMMYT
91.	TAM200/TUI	CB253	CIMMYT
92.	CROC_1/AE.SQUARROSA (205)//FCT/3/PASTOR	CB256	CIMMYT
93.	HD 2169/C591//PBW343	CB259	Pakistan
94.	V-86711TC/SH-88//CROW	CB262	Pakistan
95.	AS2002/WL711//SHAFAQ	CB266	Pakistan
96.	INQ91/YR-31	CB268	Pakistan
97.	INQ91/YR-31	CB269	Pakistan
98.	V-04179/T7 (*T. sphaerococcum*) –drought	CB276	Pakistan
99.	WBLLI*2/VIVITSI/3/T.DICOCCOMP194624/AE.SQ(409)//BCN/4/WBLL1*2/VIVTSI/5/WBLLI	CB281	CIMMYT
100.	WBLLI*2/VIVITSI/3/T.DICOCCOMP194624/AE.SQ(409)//BCN/4/WBLL1*2/VIVTSI/5/WBLLI	CB282	CIMMYT
101.	KAUZ//ALTAR84/AOS/3/MILAN/KAUZ/4/HUTES/5/T.SPELTAP1384764/6/2*KAUZ//ALTAR84/AOS	CB284	CIMMYT
102.	TOBA97/PASTOR*2//T.SPELTA P1348774	CB288	CIMMYT

Supplementary Table 1. (Cont'd.).

Sr. No.	PEDGREE Local Lines (205)	CB #	Origin
103.	T.SPELTA P1348764//INQ.91*2/TUKORU/3/WBLL1*2/TUKURU	CB290	CIMMYT
104.	V-11186	CB320	CIMMYT
105.	MUNAL #1	CB323	CIMMYT
106.	TACUPETO F2001/BRAMBLING//KIRITATI	CB326	CIMMYT
107.	ATTILA/3*BCN//BAV92/3/TILHI/5/BAV92/3/PRL/SARA//TSI/VEE#5/4/CROC_1 /AE.SQUARROSA (224)//2*OPATA	CB328	CIMMYT
108.	ROLF07*2/KIRITATI	CB329	CIMMYT
109.	FRET2/KUKUNA//FRET2/3/PARUS/5/FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TR AP//KAUZ	CB330	CIMMYT
110.	PBW343*2/KUKUNA*2//YANAC	CB332	CIMMYT
111.	TRCH//PRINIA/PASTOR	CB334	CIMMYT
112.	ACHTAR*3//KANZ/KS85-8-5/4/MILAN/KAUZ//PRINIA/3/BAV92/5/MILAN/KAUZ//PRINIA/3/BAV92	CB336	CIMMYT
113.	SOKOLL*2/TROST	CB339	CIMMYT
114.	VILLA JUAREZ F2009/SOLALA//WBLL1*2/BRAMBLING	CB341	CIMMYT
115.	NR 388	CB350	CIMMYT
116.	NR-371	CB351	Pakistan
117.	NR-379	CB354	Pakistan
118.	NR-390	CB357	Pakistan
119.	NR-403	CB362	Pakistan
120.	76377	CB369	Pakistan
121.	99108	CB372	Pakistan
122.	V-11183 –RF	CB373	Pakistan
123.	V-11365	CB375	Pakistan
124.	11B2049	CB378	Pakistan
125.	11BT004	CB381	Pakistan
126.	V-11143	CB383	Pakistan
127.	V-12284	CB385	Pakistan
128.	NW-10-1111-7	CB387	Pakistan
129.	V-11046	CB388	Pakistan
130.	NS-10	CB390	Pakistan
131.	NR 411	CB392	Pakistan
132.	V-11001	CB396	Pakistan
133.	12257	CB399	Pakistan
134.	12292	CB400	Pakistan
135.	D67.2/PARANA 66.270//AE.SQ (320)/3/CUNNINGHAM/4/VORB	CB403	Pakistan
136.	ATTILA*2/PBW65*2//HAWFINCH #1	CB411	Pakistan
137.	PFAU/SERI.1B//AMAD/3/WAXWING*2/4/MUU	CB413	Pakistan
138.	V-12253	CB420	Pakistan
139.	V-11160	CB422	Pakistan
140.	12266	CB423	Pakistan
141.	11138	CB425	Pakistan
142.	V-13005	CB427	Pakistan
143.	V-13016	CB429	Pakistan
144.	V-12130	CB433	Pakistan
145.	V-12057	CB434	Pakistan
146.	V-13241	CB435	Pakistan
147.	V-13255	CB438	Pakistan
148.	V-12066	CB440	Pakistan
149.	V-13266	CB442	Pakistan
150.	V-13270	CB443	Pakistan
151.	122557	CB450	Pakistan
152.	112095	CB452	Pakistan

Supplementary Table 1. (Cont'd.).

Sr. No.	PEDGREE Local Lines (205)	CB #	Origin
153.	12BT012	CB454	Pakistan
154.	12C027	CB456	Pakistan
155.	TW /424	CB462	Pakistan
156.	TWS12268	CB463	Pakistan
157.	MSW	CB464	Pakistan
158.	NR-429	CB465	Pakistan
159.	NR-449	CB466	Pakistan
160.	*Triticum pyrum* (V-2)	CB470	Pakistan
161.	*Triticum pyrum* (V-3)	CB471	Pakistan
162.	F6 3013 (BWP)	CB477	Pakistan
163.	088200 (mono tiller early maturity with less lodging)	CB484	Pakistan
164.	younis	CB488	Pakistan
165.	13B-3146	CB491	Pakistan
166.	TWS-12464	CB493	Pakistan
167.	NR-443	CB497	Pakistan
168.	NR-487	CB500	Pakistan
169.	14C036	CB505	Pakistan
170.	NIBGE GANDUM N	CB508	Pakistan
171.	13BT016	CB512	Pakistan
172.	14152	CB516	Pakistan
173.	14170	CB520	Pakistan
174.	14168	CB521	Pakistan
175.	13167	CB522	Pakistan
176.	14227	CB523	Pakistan
177.	13338	CB524	Pakistan
178.	AUQAB-2000	CB2	Pakistan
179.	CHAKWAL-97	CB6	Pakistan
180.	UFAQ	CB19	Pakistan
181.	CHAKWAL-86	CB5	Pakistan
182.	PUNJAB-85	CB59	Pakistan
183.	KOHISAR-95	CB12	Pakistan
184.	PARVAZ-94	CB55	Pakistan
185.	14C036	CB505	Pakistan
186.	NIBGE GANDUM N	CB508	Pakistan
187.	IV-2	CB510	Pakistan
188.	13BT016	CB512	Pakistan
189.	14152	CB516	Pakistan
190.	14170	CB520	Pakistan
191.	14168	CB521	Pakistan
192.	13167	CB522	Pakistan
193.	14227	CB523	Pakistan
194.	13338	CB524	Pakistan
195.	INQILAB 91	CB11	Pakistan
196.	AUQAB-2000	CB2	Pakistan
197.	CHAKWAL-97	CB6	Pakistan
198.	PUNJAB-96	CB60	Pakistan
199.	UFAQ	CB19	Pakistan
200.	ARRI	CB18	Pakistan
201.	KOHISTAN 97	CB50	Pakistan
202.	CHAKWAL-86	CB5	Pakistan
203.	PUNJAB-85	CB59	Pakistan
204.	KOHISAR-95	CB12	Pakistan
205.	PARVAZ-94	CB55	Pakistan

Supplementary Table 2A. Correlation matrix of 21 agro-morphological traits of 205 Pakistani spring wheat genotypes for cropping year (2017-18).

	PH	PE/L	PL	NT/P	SL	SL/S	FLL	FLW	LA	DH	DM	GFD	Y/P	B/Y	SSGW	SW	G/S	TGW	SG	WVL
PH	1																			
PE/L	0.33	1																		
PL	0.49*	0.16	1																	
NT/P	0.28	-0.04	-0.16	1																
SL	0.48*	0.04	-0.23	0.22	1															
SL/S	0.18	0.18	-0.07	0.01	0.43*	1														
FLL	0.0	-0.06	0.12	-0.01	0.13	0.03	1													
FLW	0.01	0.03	0.05	-0.00	0.03	0.07	0.43*	1												
LA	0.00	-0.01	0.11	-0.02	0.10	0.05	0.88*	0.79*	1											
DH	-0.09	-0.09	0.12	-0.07	-0.09	-0.09	0.14	0.16	0.17	1					0.07-0.0- 0-072-0.07					
DM	-0.08	-0.20	0.10	-0.02	-0.04	-0.07	0.17	0.09	0.16	0.32*	1									
GFD	-0.03	-0.15	0.02	0.01	0.01	-0.01	0.08	-0.00	0.05	-0.29	0.80*	1								
Y/P	-0.05	-0.18	-0.12	0.01	0.06	0.09	-0.00	0.03	0.01	-0.09	-0.04	0.01	1							
B/Y	0.35	0.25	-0.08	0.40*	0.27*	0.02	-0.01	-0.01	-0.01	-0.14	-0.09	-0.00	0.31*	1						
SSGW	-0.08	-0.15	0.02	0.00	-0.04	-0.03	0.11	0.10	0.12	-0.07	-0.04	0.00	0.20*	-0.15	1					
SW	-0.07	-0.18	0.00	-0.07	0.02	-0.02	0.14	0.17	0.17	-0.06	0.02	0.06	0.25*	-0.21	0.72*	1				
G/S	-0.10	-0.19	-0.02	-0.02	-0.03	-0.05	0.11	0.11	0.12	-0.00	-0.05	-0.04	0.15	-0.18	0.85*	0.60*	1			
TGW	-0.00	-0.06	0.09	-0.04	-0.00	0.03	0.12	0.08	0.12	-0.11	0.01	0.09	0.25*	-0.10	0.54*	0.52*	0.30*	1		
SG	0.101	0.065	0.150	-0.002	0.037	0.02	0.09	0.013	0.07	0.32*	0.043	-0.16	0.071	0.046	0.136	-0.15	-0.16	-0.03	1	
VWL	-0.110	0.051	0.057	-0.080	-0.045	-0.10	-0.15	-0.211	-0.22	-0.04	0.003	0.033	-0.09	0.019	-0.021	-0.09	-0.59	-0.06	-0.111	1

* Significance level alpha = 0.05

Supplementary Table 2B. Cropping Year (2018-19).

	PH	PE/L	PL	NT/P	SL	SL/S	FLL	FLW	LA	DH	DM	GFD	Y/P	B/Y	SSGW	SW	G/S	TGW	SG	WVL
PH	1																			
PE/L	0.208	1																		
PL	0.513*	0.045	1																	
NT/P	0.210	0.19	0.041	1																
SL	0.351*	0.185	0.205	0.180	1															
SL/S	0.146	0.127	-0.026	0.136	0.451*	1														
FLL	0.091	0.160	0.111	0.163	0.279	0.233	1													
FLW	-0.022	0.096	0.114	0.100	0.196	0.099	0.495*	1												
LA	0.030	0.137	0.131	0.141	0.272	0.199	0.877*	0.837*	1											
DH	0.091	0.015	0.080	0.022	0.102	0.211	0.106	0.154	0.121	1										
DM	0.032	0.094	0.078	0.094	0.118	0.239	0.101	0.240	0.187	0.579*	1									
GFD	-0.048	0.093	0.018	0.086	0.041	0.065	0.016	0.135	0.104	-0.295	0.606*	1								
Y/P	-0.101	-0.022	-0.110	-0.024	0.025	0.035	0.062	0.154	0.114	-0.085	-0.101	-0.041	1							
B/Y	0.137	0.063	0.038	0.060	0.117	0.167	0.130	0.110	0.126	-0.057	-0.048	-0.004	0.386*	1						
SSGW	-0.151	-0.062	-0.026	-0.066	0.118	0.043	0.053	0.109	0.107	-0.074	-0.069	-0.009	0.191*	0.036	1					
SW	-0.197	-0.079	-0.031	-0.084	0.145	0.058	0.011	0.144	0.099	-0.061	-0.011	0.045	0.250*	0.067	0.724*	1				
G/S	-0.220	-0.086	-0.062	-0.092	0.065	0.054	-0.006	0.068	0.044	-0.011	-0.007	0.000	0.147	0.001	0.862*	0.605*	1			
TGW	0.090	-0.028	0.043	-0.030	0.208	0.015	-0.079	0.006	-0.038	-0.121	-0.125	-0.030	0.253*	0.105	0.531*	0.522*	0.302*	1		
SG	0.101	0.065	0.150	-0.002	0.037	0.020	0.096	0.013	0.073	0.329*	0.043	-0.161	0.071	0.046	0.136	-0.151	-0.164	-0.036	1	
VWL	-0.110	0.051	0.057	-0.080	-0.045	-0.103	-0.159	-0.211	-0.220	-0.048	0.003	0.033	-0.098	0.019	-0.021	-0.090	-0.-59	-0.065	-0.11	1

* Significance level alpha = 0.05

Supplementary Table 2C. (Control).

Control	PH	PE/L	PL	NT/P	SL	SL/S	FLL	FLW	LA	DH	DM	GFD	Y/P	B/Y	SSGW	SW	G/S	TGW	SG	VWL
PH	1																			
PE/L	0.139	1																		
PL	0.55*	0.14	1																	
NT/P	0.289	-0.107	-0.006	1																
SL	0.476*	0.205	0.057	-0.009	1															
SL/S	0.180	0.044	0.089	0.012	0.47*	1														
FLL	-0.041	0.111	0.085	-0.071	0.279	0.090	1													
FLW	0.020	-0.004	0.005	0.090	0.141	0.077	0.435*	1												
LA	-0.047	0.131	0.073	-0.083	0.270	0.036	0.874*	0.773*	1											
DH	-0.098	0.078	0.133	-0.073	0.101	-0.091	0.105	-0.011	0.124	1										
DM	-0.136	0.078	0.054	-0.026	0.118	-0.097	0.101	0.040	0.180	0.575*	1									
GFD	-0.073	0.021	-0.066	0.035	0.042	-0.021	0.016	0.059	0.092	-0.29	0.606*	1								
Y/P	-0.071	-0.110	-0.139	-0.001	0.025	0.095	0.062	0.087	0.123	-0.083	-0.101	-0.045	1							
B/Y	0.106	0.011	-0.058	0.017	0.102	0.092	0.117	0.102	0.125	-0.062	-0.065	-0.021	0.36*	1						
SSGW	-0.074	-0.026	-0.124	0.001	0.118	-0.045	0.053	-0.118	0.110	-0.074	-0.069	-0.009	0.191*	0.033	1					
SW	-0.070	-0.031	-0.170	-0.078	0.145	-0.027	0.011	-0.075	0.103	-0.061	-0.011	0.044	0.25*	0.065	0.724*	1				
G/S	-0.100	-0.062	-0.106	-0.018	0.065	-0.057	-0.006	-0.146	0.048	-0.010	-0.007	-0.001	0.147	-0.004	0.862*	0.605*	1			
TGW	0.008	0.027	-0.107	-0.003	0.223	0.057	-0.065	0.308*	-0.054	-0.132	-0.128	-0.024	0.25*	0.125	0.453*	0.456*	0.231*	1		
SG	0.101	0.065	0.150	-0.002	0.037	0.020	0.096	0.013	0.073	0.329*	0.043	-0.161	0.071	0.046	0.136	-0.151	-0.164	-0.036	1	
VWL	-0.110	0.051	0.057	-0.080	-0.045	-0.103	-0.159	-0.211	-0.220	-0.048	0.003	0.033	-0.098	0.019	-0.021	-0.090	-0.-59	-0.065	-0.11	1

* Significance level alpha = 0.05

Supplementary Table 3. Response genotypes on GLR scale during both growing seasons 2017-18 and 2018-19.

CB #	Score	Year 1	Score	Year 2	CB #	Score	Year 1	Score	Year 2	CB #	Score	Year 1	Score	Year 2	CB #	Score	Year 1	Score	Year 2	CB #	Score	Year 1	Score	Year 2	CB #	Score	Year 1	Score	Year 2
CB3	1	R	2	MR	CB99	5	MR	6	MS	CB186	8	MS	5	MR	CB266	6	MS	6	MS	CB381	3	MR	5	MR	CB463	8	MS	6	MS
CB4	0	R	1	R	CB100	8	MS	8	MS	CB188	8	MS	5	MR	CB268	7	MS	3	MR	CB383	0	R	2	MR	CB464	8	MS	6	MS
CB8	3	MR	4	MR	CB103	9	S	9	S	CB195	8	MS	5	MR	CB269	8	MS	9	S	CB385	4	MR	5	MR	CB465	5	MR	5	MR
CB11	2	MR	4	MR	CB104	6	MS	7	MS	CB196	5	MR	7	MS	CB276	2	MR	7	MS	CB387	1	R	1	R	CB466	8	MS	8	MS
CB14	0	R	2	MR	CB106	4	MR	4	MR	CB198	7	MS	7	MS	CB281	3	MR	3	MR	CB388	2	MR	2	MR	CB470	0	R	0	R
CB15	0	R	1	R	CB107	5	MR	5	MR	CB200	6	MS	4	MR	CB282	6	MS	9	S	CB390	4	MR	6	MS	CB471	8	MS	8	MS
CB17	0	R	3	MR	CB115	0	R	3	MR	CB202	5	MR	6	MS	CB284	8	MS	8	MS	CB392	5	MR	5	MR	CB477	8	MS	8	MS
CB20	0	R	2	MR	CB117	4	MR	4	MR	CB203	0	R	0	R	CB288	7	MS	5	MR	CB393	5	MR	7	MS	CB484	5	MR	7	MS
CB23	5	MR	7	MS	CB120	6	MS	6	MS	CB208	5	MR	6	MS	CB290	8	MS	8	MS	CB396	6	MS	8	MS	CB488	8	MS	8	MS
CB29	8	MS	9	S	CB121	6	MS	4	MR	CB210	0	R	2	MR	CB315	8	MS	4	MR	CB399	7	MS	9	S	CB491	0	R	5	MR
CB30	2	MR	3	MR	CB123	5	MR	3	MR	CB212	0	R	1	R	CB318	7	MS	7	MS	CB400	5	MR	5	MR	CB493	4	MR	4	MR
CB37	3	MR	4	MR	CB124	5	MR	5	MR	CB214	7	MS	7	MS	CB320	8	MS	6	MS	CB403	5	MR	1	R	CB497	6	MS	6	MS
CB42	8	MS	8	MS	CB126	6	MS	4	MS	CB216	5	MR	4	MR	CB323	8	MS	8	MS	CB411	6	MS	6	MS	CB498	7	MS	7	MS
CB46	8	MS	8	MS	CB128	8	MS	8	MS	CB220	1	R	4	MR	CB326	5	MR	9	S	CB413	3	MR	7	MS	CB500	5	MR	5	MR
CB47	6	MS	7	MS	CB129	8	MS	8	MS	CB224	0	R	4	MR	CB328	8	MS	8	MS	CB420	8	MS	8	MS	CB505	4	MR	8	MS
CB50	7	MS	7	MS	CB130	3	MR	5	MR	CB228	5	MR	5	MR	CB329	7	MS	3	MR	CB422	9	S	9	S	CB508	7	MS	9	S
CB56	0	R	2	MR	CB131	8	MS	8	MS	CB232	4	MR	6	MS	CB330	7	MS	5	MR	CB423	3	MR	6	MS	CB510	5	MR	5	MR
CB57	5	MR	6	MS	CB133	0	R	3	MR	CB234	2	MR	5	MR	CB332	5	MR	3	MR	CB425	0	R	0	R	CB512	3	MR	3	MR
CB60	6	MS	6	MS	CB138	5	MR	5	MR	CB236	3	MR	5	MR	CB334	6	MS	2	MR	CB427	3	MR	3	MR	CB516	2	MR	6	MR
CB61	7	MS	8	MS	CB142	6	MS	7	MS	CB238	4	MR	6	MS	CB336	5	MR	1	R	CB429	4	MR	4	MR	CB520	0	R	0	R
CB64	0	R	0	R	CB146	7	MS	7	MS	CB239	5	MR	5	MR	CB339	7	MS	9	S	CB432	5	MR	5	MR	CB521	0	R	0	R
CB65	8	MS	7	MS	CB147	4	MR	6	MS	CB240	0	R	0	R	CB341	2	MR	7	MS	CB433	0	R	0	R	CB522	0	R	1	R
CB66	5	MR	5	MR	CB148	1	R	1	R	CB241	0	R	5	MR	CB345	5	MR	9	S	CB434	0	R	0	R	CB523	5	MR	5	MR
CB69	7	MS	6	MS	CB150	0	R	3	MR	CB242	0	R	0	R	CB349	4	MR	5	MR	CB435	4	MR	4	MR	CB524	4	MR	4	MR
CB73	3	MR	3	MR	CB154	8	MS	8	MS	CB244	1	R	4	MR	CB350	8	MS	9	S	CB438	3	MR	3	MR	CB11	7	MS	7	MS
CB74	5	MR	6	MS	CB163	8	MS	7	MS	CB247	8	MS	6	MS	CB351	9	S	8	MS	CB440	3	MR	6	MS	CB2	5	MR	5	MR
CB75	7	MS	7	MS	CB165	2	MR	2	MR	CB248	1	R	1	R	CB354	6	MS	3	MR	CB442	6	MS	6	MS	CB6	4	MR	4	MR
CB76	0	R	3	MR	CB167	3	MR	3	MR	CB249	8	MS	9	S	CB357	4	MR	0	R	CB443	4	MR	4	MR	CB60	4	MR	4	MR
CB80	8	MS	9	S	CB171	5	MR	3	MR	CB251	5	MR	3	MR	CB362	2	MR	6	MS	CB450	4	MR	4	MR	CB19	3	MR	7	MS
CB82	5	MR	5	MR	CB172	8	MS	8	MS	CB252	5	MR	8	MS	CB369	7	MS	7	MS	CB452	8	MS	8	MS	CB18	0	R	0	R
CB83	6	MS	7	MS	CB174	6	MS	5	MR	CB253	7	MS	4	MR	CB372	6	MS	6	MS	CB454	6	MS	6	MS	CB50	5	MR	5	MR
CB85	2	MR	2	MR	CB176	8	MS	8	MS	CB256	8	MS	9	S	CB373	0	R	3	MR	CB456	8	MS	8	MS	CB5	6	MS	6	MS
CB94	5	MR	6	MS	CB179	2	MR	2	MR	CB259	8	MS	8	MS	CB375	7	MS	2	MR	CB458	5	MR	7	MS	CB59	7	MS	9	S
CB96	8	MS	8	MS	CB182	5	MR	6	MS	CB262	5	MR	1	R	CB378	0	R	1	R	CB462	8	MS	6	MS	CB12	7	MS	7	MS

Supplementary Table 4. Heat susceptibility index of year 2017-18 and 2018-19.

CB. No	HSI-2018		HSI-2019		CB. No	HSI-2018		HSI-2019		CB. No	HSI-2018		HSI-2019		CB. No	HSI-2018		HSI-2019		CB. No	HSI-2018		HSI-2019		CB. No	HSI-2018		HSI-2019	
CB3	1.20	HS	1.26	HS	CB99	1.86	HS	1.06	HS	CB186	1.69	HS	2.70	HS	CB266	0.65	MT	0.79	MT	CB381	1.11	HS	1.20	HS	CB463	0.95	MT	0.75	MT
CB4	1.22	HS	1.11	HS	CB100	0.92	MT	0.98	MT	CB188	1.58	HS	1.10	HS	CB268	0.82	MT	0.80	MT	CB383	0.35	MT	0.84	MT	CB464	0.59	MT	0.81	MT
CB8	0.68	MT	0.79	MT	CB103*	0.30	HHT	0.37	HHT	CB195	0.82	MT	0.75	MT	CB269	1.55	HS	1.97	HS	CB385	1.43	HS	1.00	HS	CB465	0.79	MT	0.94	MT
CB11*	0.23	HHT	0.34	HHT	CB104	1.96	HS	1.93	HS	CB196	2.32	HS	1.13	HS	CB276	1.70	HS	0.52	HS	CB387	0.54	MT	0.79	MT	CB466	1.11	HS	1.09	HS
CB14	1.44	HS	1.58	HS	CB106	1.40	HS	0.68	MT	CB198	1.07	HS	1.44	HS	CB281	0.58	MT	0.99	MT	CB388	0.66	MT	0.76	MT	CB470	1.94	HS	1.03	HS
CB15	1.34	HS	1.14	HS	CB107	0.77	MT	0.66	HS	CB200	0.73	MT	0.77	MT	CB282	1.69	HS	1.88	HS	CB390	1.10	HS	1.20	HS	CB471	1.15	HS	1.97	HS
CB17	1.50	HS	1.92	HS	CB115	1.20	HS	1.13	HS	CB202	1.27	HS	1.89	HS	CB284	0.82	MT	0.95	MT	CB392	1.93	HS	1.00	HS	CB477	1.15	HS	1.05	HS
CB20	1.56	HS	2.61	HS	CB117	1.73	HS	1.15	HS	CB203	0.70	MT	0.84	MT	CB288	0.57	MT	0.70	MT	CB393	0.56	MT	0.89	MT	CB484	0.99	HS	0.83	HS
CB23	0.94	MT	0.97	MT	CB120	1.55	HS	1.80	HS	CB208	1.74	HS	1.96	HS	CB290	1.74	HS	1.37	HS	CB396	0.52	MT	0.95	MT	CB488	1.06	HS	1.80	HS
CB29	1.82	HS	1.07	HS	CB121	1.04	HS	1.31	HS	CB210	0.73	MT	0.80	MT	CB315	1.07	HS	1.40	HS	CB399	0.79	MT	0.81	MT	CB491	0.75	MT	0.81	MT
CB30	2.06	HS	1.80	HS	CB123	1.28	HS	1.92	HS	CB212	2.65	HS	1.70	HS	CB318	0.86	MT	0.84	MT	CB400*	0.42	HHT	0.37	HHT	CB493	0.78	MT	0.84	MT
CB37	1.88	HS	1.93	HS	CB124	0.78	MT	0.96	MT	CB214	1.12	HS	1.10	HS	CB320	1.86	HS	1.42	HS	CB403*	0.45	HHT	0.31	HHT	CB497	0.75	MT	0.90	MT
CB42	0.97	MT	0.82	MT	CB126	0.77	MT	0.82	MT	CB216*	0.34	HHT	0.25	HHT	CB323	0.73	MT	0.75	MT	CB411	0.98	MT	0.76	MT	CB498	1.96	HS	1.04	HS
CB46	1.22	HS	1.06	HS	CB128	1.97	HS	1.81	HS	CB220	0.98	MT	0.61	MT	CB326	1.13	HS	1.15	HS	CB413	1.55	HS	1.06	HS	CB500	0.79	MT	0.90	MT
CB47	1.34	HS	1.41	HS	CB129	1.78	HS	1.13	HS	CB224	1.67	HS	1.27	HS	CB328	1.52	MT	1.99	HS	CB420	0.52	MT	0.76	MT	CB505	1.13	HS	1.13	HS
CB50	1.91	HS	1.08	HS	CB130	1.35	HS	1.24	HS	CB228*	0.21	HHT	0.32	HHT	CB329	1.04	HS	1.03	HS	CB422	1.34	HS	1.09	HS	CB508	2.10	HS	2.50	HS
CB56	0.63	MT	0.76	MT	CB131	1.60	HS	1.01	HS	CB232	1.63	HS	1.95	HS	CB330	1.86	HS	1.02	HS	CB423	0.51	MT	0.82	MT	CB510	0.81	MT	0.73	MT
CB57	0.61	MT	0.70	MT	CB133	0.70	MT	0.67	MT	CB234	0.80	MT	0.79	MT	CB332	0.82	MT	0.81	MT	CB425	1.10	HS	1.13	HS	CB512	0.62	MT	0.72	MT
CB60	1.15	HS	1.18	HS	CB138	2.35	HS	1.75	HS	CB236	0.81	MT	0.97	MT	CB334	0.13	HS	0.17	HS	CB427	1.14	HS	1.15	HS	CB516	0.63	MT	0.54	MT
CB61	1.22	HS	1.20	HS	CB142	0.65	MT	0.89	MT	CB238	0.80	MT	0.74	MT	CB336	1.44	HS	1.09	HS	CB429	0.69	MT	0.74	MT	CB520	0.63	MT	0.88	MT
CB64	1.89	HS	1.17	HS	CB146	0.85	MT	0.94	MT	CB239	0.67	MT	0.71	MT	CB339	0.66	MT	0.77	MT	CB432	1.12	HS	1.16	HS	CB521	1.84	HS	2.14	HS
CB65	0.94	MT	0.97	MT	CB147	1.29	HS	1.01	HS	CB240	0.21	HS	1.21	HS	CB341	1.66	HS	1.33	HS	CB433	0.99	MT	1.37	HS	CB522	0.97	MT	0.79	MT
CB66	1.99	HS	1.17	HS	CB148	1.48	HS	1.83	HS	CB241	0.83	MT	0.92	MT	CB345	0.81	MT	0.89	MT	CB434	1.07	HS	0.58	MT	CB523	0.65	MT	0.59	MT
CB69	1.91	HS	1.15	HS	CB150	0.94	MT	0.68	MT	CB242	0.80	MT	0.88	MT	CB349	0.63	MT	0.82	MT	CB435	1.11	HS	1.20	HS	CB524	1.00	HS	1.89	HS
CB73	1.03	HS	1.05	HS	CB154	1.11	HS	1.52	HS	CB244	1.69	HS	1.32	HS	CB350	0.53	MT	0.72	MT	CB438	0.54	MT	0.80	MT	CB11	0.75	MT	0.57	MT
CB74	0.88	MT	0.95	MT	CB163	1.10	HS	1.50	HS	CB247	2.29	HS	1.03	HS	CB351	0.79	MT	0.94	MT	CB440	1.35	HS	1.03	HS	CB2	0.65	MT	0.61	MT
CB75	1.13	HS	1.15	HS	CB165	0.60	MT	0.93	MT	CB248	1.59	HS	1.23	HS	CB354	1.44	HS	1.01	HS	CB442	1.18	HS	0.80	MT	CB6	1.13	HS	1.13	HS
CB76	1.28	HS	1.06	HS	CB167	1.37	HS	1.06	HS	CB249	0.99	MT	0.86	MT	CB357	1.45	HS	1.01	HS	CB443	1.11	HS	1.98	HS	CB60	0.54	MT	0.58	MT
CB80	0.99	MT	0.88	MT	CB171	0.82	MT	0.74	MT	CB251	1.14	HS	1.11	HS	CB362	1.52	HS	1.26	HS	CB450	1.67	HS	1.04	HS	CB19*	0.24	HHT	0.34	HHT
CB82	1.55	HS	1.83	HS	CB172	0.64	MT	0.63	MT	CB252	1.86	MS	1.37	HS	CB369	1.56	HS	1.10	HS	CB452	1.10	HS	0.37	HS	CB18	0.71	MT	0.55	MT
CB83	1.84	HS	1.17	HS	CB174	1.48	HS	1.70	HS	CB253	0.99	MT	0.95	MT	CB372	1.42	HS	1.10	HS	CB454	1.88	HS	1.24	HS	CB50	0.53	MT	0.60	MT
CB85	1.32	HS	1.27	HS	CB176	0.83	MT	0.67	MT	CB256	0.97	MT	0.87	MT	CB373*	0.20	HHT	0.17	HHT	CB456	2.35	HS	0.85	MT	CB5	0.16	MT	0.12	MT
CB94	1.07	HS	1.07	HS	CB179	1.98	HS	1.15	HS	CB259	1.52	HS	1.09	HS	CB375	0.96	MT	0.87	MT	CB458	1.92	HS	0.95	MT	CB59	0.55	MT	0.60	MT
CB96	0.80	MT	0.97	MT	CB182	0.98	MT	0.89	MT	CB262	1.03	HS	1.26	HS	CB378	1.10	HS	1.88	HS	CB462	1.38	HS	1.02	HS	CB12	0.56	MT	0.58	MT

Supplementary Table 5. Kernel weight reduction (KWR) of year 2017-18 and 2018-19.

CB No	%KWR 2018	%KWR 2019	CB No	%KWR 2018	%KWR 2019	CB No	%KWR 2018	%KWR 2019	CB No	%KWR 2018	%KWR 2019	CB No	%KWR 2018	%KWR 2019	CB No	%KWR 2018	%KWR 2019
CB3**	38.129	21.036	CB99	17.06	9.41	CB186	18.65	10.29	CB266	18.26	10.08	CB381	20.95	11.56	CB463	21.46	11.84
CB4	20.949	11.557	CB100	16.49	9.10	CB188	18.65	10.29	CB268	17.00	9.38	CB383	16.74	9.24	CB464	18.26	10.08
CB8	21.048	11.612	CB103*	11.95	8.80	CB195	30.08	16.59	CB269	19.91	10.98	CB385	19.30	10.65	CB465	17.82	9.83
CB11*	10.930	8.237	CB104	16.61	9.17	CB196	19.13	10.56	CB276**	41.73	23.02	CB387	19.22	10.60	CB466	21.46	11.84
CB14	21.354	11.781	CB106	18.57	10.25	CB198**	51.46	28.39	CB281	21.05	11.61	CB388	17.33	9.56	CB470	17.54	9.68
CB15	18.416	10.160	CB107	21.15	11.67	CB200	16.49	9.10	CB282	28.71	15.84	CB390	19.39	10.69	CB471	19.56	10.79
CB17	17.197	9.487	CB115**	40.96	22.60	CB202	18.34	10.12	CB284	18.42	10.16	CB392	17.68	9.75	CB477	17.68	9.75
CB20	20.000	11.034	CB117	18.49	10.20	CB203	20.65	11.40	CB288	21.25	11.72	CB393	21.15	11.67	CB484	17.75	9.79
CB23	20.091	11.084	CB120	21.99	12.13	CB208	20.46	11.29	CB290**	35.10	19.36	CB396	19.13	10.56	CB488	18.73	10.33
CB29	24.143	10.193	CB121	19.13	10.56	CB210	20.95	11.56	CB315	20.46	11.29	CB399	18.42	10.16	CB491	17.89	9.87
CB30	21.883	12.073	CB123	26.32	14.52	CB212	17.97	9.91	CB318	19.73	10.89	CB400*	9.40	13.50	CB493	20.65	11.40
CB37	20.091	11.084	CB124	26.63	14.69	CB214	18.81	10.38	CB320	17.97	9.91	CB403*	11.45	12.52	CB497	16.74	9.24
CB42	18.039	9.952	CB126	17.00	9.38	CB216*	7.19	9.38	CB323**	35.38	19.52	CB411	18.73	10.33	CB498	17.54	9.68
CB46	20.949	11.557	CB128	20.65	11.40	CB220	21.67	11.95	CB326	19.47	10.74	CB413	18.57	10.25	CB500	20.28	11.19
CB47	20.091	11.084	CB129	15.78	8.71	CB224	17.47	9.64	CB328	35.96	19.84	CB420	17.89	9.87	CB505	21.77	12.01
CB50	23.266	12.836	CB130	29.28	16.15	CB228*	10.95	8.80	CB329	18.73	10.33	CB422**	32.52	17.94	CB508**	42.95	23.70
CB56	27.461	15.150	CB131	29.88	16.48	CB232	19.13	10.56	CB330	29.48	16.26	CB423	22.55	12.44	CB510	19.56	10.79
CB57	18.416	10.160	CB133	17.68	9.75	CB234	19.56	10.79	CB332	24.29	13.40	CB425	20.18	11.13	CB512	18.65	10.29
CB60	24.025	13.255	CB138	21.88	12.07	CB236	19.47	10.74	CB334**	38.13	21.04	CB427	17.33	9.56	CB516	19.73	10.89
CB61	19.557	10.790	CB142	22.90	12.64	CB238	16.49	9.10	CB336	16.49	9.10	CB429	16.93	9.34	CB520	18.04	9.95
CB64	19.217	10.602	CB146	16.93	9.34	CB239	17.61	9.71	CB339	21.67	11.95	CB432	23.27	12.84	CB521	22.55	12.44
CB65	24.290	13.401	CB147	21.77	12.01	CB240	16.37	9.03	CB341	19.56	10.79	CB433**	33.76	18.62	CB522	22.55	12.44
CB66	17.678	9.753	CB148	22.55	12.44	CB241	19.91	10.98	CB345	25.12	13.86	CB434	23.14	12.77	CB523	18.73	10.33
CB69	16.614	9.166	CB150	18.26	10.08	CB242**	38.13	21.04	CB349	17.26	9.52	CB435	18.19	10.03	CB524	17.68	9.75
CB73	21.562	11.896	CB154	20.28	11.19	CB244	26.63	14.69	CB350	20.56	11.34	CB438	19.56	10.79	CB11	21.46	11.84
CB74	23.023	12.702	CB163	22.55	12.44	CB247	19.91	10.98	CB351	26.63	14.69	CB440	16.87	9.31	CB2	18.89	10.42
CB75	26.160	14.432	CB165	23.02	12.70	CB248	20.28	11.19	CB354	16.61	9.17	CB442	18.57	10.25	CB6	21.46	11.84
CB76	19.051	10.510	CB167	19.13	10.56	CB249**	28.72	33.96	CB357	17.26	9.52	CB443	24.84	13.70	CB60	22.33	12.32
CB80	19.732	10.886	CB171	17.26	9.52	CB251	18.57	10.25	CB362	19.22	10.60	CB450	18.26	10.08	CB19*	9.19	12.38
CB82	18.339	10.118	CB172	18.73	10.33	CB252	20.95	11.56	CB369	18.04	9.95	CB452	17.20	9.49	CB18**	32.52	17.94
CB83	26.474	14.605	CB174	20.65	11.40	CB253	22.55	12.44	CB372	19.91	10.98	CB454	21.99	12.13	CB50	22.90	12.64
CB85	19.051	10.510	CB176	18.73	10.33	CB256	22.33	12.32	CB373*	15.29	8.44	CB456	19.22	10.60	CB5	21.99	12.13
CB94	15.671	8.646	CB179	20.46	11.29	CB259	18.81	10.38	CB375	26.63	14.69	CB458	23.27	12.84	CB59	19.91	10.98
CB96	23.767	13.112	CB182	18.73	10.33	CB262**	36.86	20.33	CB378	17.68	9.75	CB462	20.65	11.40	CB12	17.33	9.56