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STABILITY OF SOME HEAT TOLERANT TRAITS IN A DOUBLE HAPLOID POPULATION OF WHEAT (*TRITICUM AESTIVUM* L.) IN THE EASTERN GANGETIC PLAINS OF INDIA

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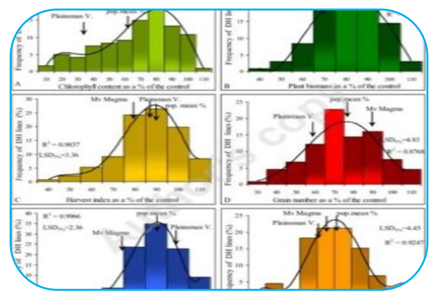
ABSTRACT:



The high temperature during grain filling stage causes significant yield losses to wheat in many parts of the world including south Asia. One hundred twenty doubled haploid (DH) wheat lines derived from the cross Sokol/Krichauff, were grown under late sown condition during four consecutive crop seasons (2007-2011) at Banaras Hindu University, Varanasi. The objective of this experiment was to assess the most contributing trait for terminal heat tolerance and to identify superior DH lines. In this regard a significant correlation was observed among grain yield (GY), grain filling duration (GFD), and canopy temperature (CT). The pooled analysis of variance revealed highly significant differences for genotypes, environment, and genotype × environment interaction for the three traits. This indicated considerable diversity among DH lines with a significant degree of environmental influence. On the basis of GY three lines were significantly superior while rest of other lines were found to be at par to parent. On the basis of stability parameter 3 DH lines were found to be stable for GFD, 5 for GY and 11 for CT.

KEY WORDS: doubled haploid (DH) , grain yield (GY), grain filling duration (GFD).

INTRODUCTION

To ensure the food security in south Asia, wheat (*Triticum aestivum* L.) is of significant importance [1]. Among the major factors threatening wheat production in south Asia, terminal heat is of utmost importance [2]. Terminal heat stress is very crucial abiotic stress that can be problem in 40% of the irrigated wheat growing areas of the developing world which covers ~36 m ha including the eastern Gangetic plains (EGP) [3, 4, 5]. In about five million hectares of eastern Gangetic Plains (EGP) of India, wheat sowing gets delayed to the second fortnight of December resulting in high yield losses caused by significant shrinkage in the period available for the growth and development of wheat crop [6, 7]. The unusual warming trends during grain filling period continues to be a cause of serious worry for wheat growers and researchers [8], therefore the need becomes warranted for the breeders to think seriously for developing heat tolerant locally adaptable wheat varieties that performs good, without compromising the yield under stress conditions.

Yield reduction in wheat under heat stress could be attributed to various factors *i.e.*, accelerated phasic development [9], accelerated senescence [10], increased respiration [11], reduced photosynthesis [12] and inhibition of starch synthesis in developing kernels [13], high temperatures during early crop development, particularly after anthesis [14]. However, it has been suggested that there is substantial scope for the improvement in productivity under unfavorable environments characterized by significant presence of abiotic stress that includes high temperature and terminal heat stress [15, 2].

Genotype × environment (GE) interactions are of immense importance in the development and evaluation of plant varieties since they reduce the genotypic stability values under diverse environments [16]. The concept of stability has been defined in several ways through several biometrical methods including univariate and multivariate ones [17]. However, the most widely used one is the regression method, based on regressing the mean value of each genotype on the environmental index or marginal means of environments [18]. [19] proposed a method for measuring stability that was later improved [20]. Stability parameters have earlier been investigated in wheat varieties [21, 22]. Environmental conditions are known to have significant influence on yield of wheat. According to [23], in the presence of G × E interactions, stability parameters are estimated to determine the superiority of individual genotypes across the range of environments. Therefore, evaluation of genotypes for stability of performance under varying environmental conditions for yield has become an essential part of breeding programmes.

The present study was carried out with the purpose of analyzing stability of performance of wheat DH lines under heat stressed environments in the EGP and to find out heat tolerant DH lines having value for the future.

MATERIAL AND METHODS

One hundred twenty double haploid (DH) lines generated in SARDI Australia from two parents Sokol and Krichauff were the study population . Both parent were heat adapted. The check used was DBW-14. The experiment was conducted at Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi (25.18^0 N latitude; 83.03^0 E longitudes, 75.7 m above mean sea level, soil pH 7.4) which falls under the EGP of India. Wheat in the EGP is invariably exposed of high temperature particularly at the time of its growth and development and at the end of the wheat season in the EGP of India [24] which is considered as a heat-prone mega-environment (ME5) [25]). The experiments were planted in four consecutive seasons 2007-08, 2008-09, 2009-10, 2010-11 under late sown condition (3rd week of December) to represent heat stressed environments.

Daily temperatures (Fig. 1) indicate that the late sown experiment was heat stressed. All the plantings were conducted under irrigated conditions with four irrigations applied during the entire experimental period. The trial was sown in a Randomized Complete Block Design with 6 row plots of 3 m length and 25 cm spacing between the rows (4.5 m^2) in 3 replications each with four blocks having 30 lines with a check (HUW 468) after every 5th line to serve as covariate in ANOVA analysis. The covariate values for each line were determined as the average of the checks lying on its two sides i.e., at the beginnings/ends of the blocks. However, where the lines had the checks at only one side of them, the value of the check nearest to them were used.

The normal prescribed input management was followed i.e., the fertilizer was applied as per the recommendations for the EGP i.e., 120 kg of N, 60 kg of P_2O_5 and 40 kg of K_2O per hectare in each year. While P_2O_5 and K_2O were applied during sowing, the N application was split in 3 doses; during sowing and one each after the 1st and 2nd irrigations. To control the disease spread the crop was protected from spot blotch and leaf rust (two of the most serious fungal diseases of wheat in EGP) by applying the fungicide Tilt (Propiconazole); @ 625 g a.i./ha) at two growth stages GS 54 and 69 (Zadoks et al. 1974) [25]. The plots were kept weed free by applying pre-emergence herbicide Pendimethalin (@ 1000 g/ha in 500-600 liters of water/ha at 3 days after sowing) and Sulfosulfuron (@ 25.0 g/ha in 250-300 litres of water/ha was sprayed at 25 days after sowing).

The traits recorded were grain yield (GY), grain filling duration (GFD), canopy temperature (CT), date of anthesis and physiological maturity (75% of the plants mature per plot) both for the DH population as well as parental genotypes. The days to anthesis was subtracted from days to physiological maturity to obtain the GFD of each line.

The CT was recorded at three growth stages of crop viz., heading (GS55), anthesis (GS65) and early dough (GS83) [26] on bright sunny days between 1200h and 1400h at 7 day intervals. For this measurement a hand held infra red thermometer (Mikron Infrared Inc. NJ, USA) was used holding at an angle of 30^0 to the horizontal plane, one meter away from the edge of the plot and approximately 50 cm above the crop, giving a canopy view of 10 cm × 25cm.

RESULTS:

The results of combined analysis of variance based on Eberhart and Russell stability method, 1966 [20] are presented in Table-1. The mean square due to genotypes and environments for all the traits was found to be significant, suggesting the existence of considerable variation among genotypes as well as environments. The Genotype × environment interaction was found to be significant for all the three characters indicating that they were highly influenced by the changes in environments. The variation due to pooled deviation was significant for all the characters suggesting that performance of different DH lines fluctuated **s**ignificantly from their respective linear path of response to environment. The daily temperatures during the experimental period (Fig. 1) indicated that temperatures showed an increasing trend after mid February when the crop was at the grain filling stage and there was significant difference in the expression of the traits under heat stressed conditions. The heritability (broad sense) also varied for the all the traits. The heritability for GY, GFD and CT, was 65%, 55% and 40 % respectively (Table 1).

Substantial variation was noted in DH mean for GY, GFD, and CT in each year with much variation in DH range along with both parents (Table 2). There was positive significant correlation (0.319*) between GFD and GY and negative correlation (-0.149) between GY and CT (Table 3)

On the basis of GY, the top 20 lines were selected in which three lines (no. 40, 41 and 12) were significantly superior while rest of other lines were found to be at par to parents interestingly the line no. 40 was also significantly superior to the check used DBW-14 (Table 4). Twenty best lines were observed and listed in Table 5 on the basis of better results on GFD and CT as compared to the parents. Results of stability analysis and bi, S^2di with mean of all traits [20] are presented in Table-6.

The results of stability analysis indicated that in case of GY, 29 lines had higher mean than the parents, while 97 were significantly stable showing bi=1, and 11 lines had non significant value of S^2di. It was found that for GFD 33 DH lines had high mean value than parents; however 108 lines had unit bi value while 28 had non significant S^2di value. Finally for CT, 86 lines had mean higher than best parent while 88 lines had unit bi value and 107 lines were having non significant S^2di value. All the genotypes were found to be showing positive bi value for the traits studied (data not shown).

When the parameters higher mean than parent, bi=1 and non significant S^2di were considered together, 5 DH lines (no. 3, 5, 22, 35, 106) were found to be stable for GY. Three lines (no. 39, 63, and 69) were found stable for GFD and 13 lines (no.10, 11, 15, 16, 18, 20, 21, 22, 23, 24, 63, 69, 106) were stable for CT (Table 6, Fig 2). Line numbers 22 and 106 were found to be stable for two traits GY and CT while considering all the three parameters of stability for four environments studied. Line no. 3, 5, 22 and 106 were observed to be superior or at par in yield as compared to the parents and these lines also showed stability for yield in terms of all parameters [20].

DISCUSSION

The growing constraint on wheat cultivation can be understood by the fact that a decade ago, 40°C before March 30 was uncommon in the eastern Gangetic plains, but now it is so frequent and often even before March 30, with 40°C being recorded, on average, around one week earlier than previously observed [24].

Current estimates indicate that in India alone around among 13.5 mha of wheat area is heat stressed [24] which may grow higher in coming next decade [27, 28], genetic variation for heat tolerance in wheat cultivars is well established [29, 4]. The significant mean square due to genotypes and environment for GY, GFD and CT seen in the result, suggested the existence of considerable variation among the genotypes for the traits (Table 1). A purely statistical analysis, was proposed [30] that was later used by [19] and [20].

Estimation of correlation coefficient all the traits could be considered the best and most desirable in any experiment. As evident, all the other traits *i.e.,* GFD are yield contributing traits and had positive correlation with each other. Negative correlation of GY with CT indicated the importance of low value of CT. Heritability for GFD was lower than that reported earlier [31] who reported a broad-sense heritability of 80%. Similarly the moderate heritability for CT found in our experiments was in accordance with the earlier results of [4]. The high heritable traits *viz.,* GY, GFD and CT could be considered desirable to use as selection criterion for heat stress tolerance. Higher H^2 indicates genotype x environment interactions to be low, although G x E was significant for all the traits under late sown conditions. Exposure to higher than optimal temperatures reduces yield and decreases the quality of wheat [32, 33, 34]. While CT shows a strong and reliable association with yield under drought and heat stress and is used in wheat breeding to select for yield. Canopy temperatures are determinative as CTD for heat tolerance [35, 36]. Since leaf temperature is depressed below air temperature when water evaporates, CT is an indirect measure of the transpiration at the whole-crop level [4] and plant water status [37].

As explained by [19], linear (bi) and non- significant (S^2di) should be considered while judging the phenotypic stability of a variety. They also emphasized that an ideal variety should have high mean performance better than best parent, “b” value near to unity and S^2di close to zero. When considering all the parameters of stability together it was evident that there were lines (Table 6) that were less sensitive to environments and this is desirable for all the traits i.e. GY, GFD and CT.

Yield stability has been defined as a measure of variation between potential and actual yield of genotypes across different environments [38]. The yield stability of a cultivar is influenced by the genotype of individual plants and the genetic constitution of the lines [39].

All the genotype mention in Table 6 showed non significant S^2di value for all the traits suggesting these genotypes to be relatively stable under environmental fluctuations. It has also been reported that the consideration should be given to those varieties, which produced higher mean yield with small deviation from regression coefficient and regression coefficient equal to one [40].

The superior performance of some of the DH lines appears to suggest scope of obtaining transgressive segregants even in doubled haploid populations that is due to additive effects of favorable genes present in the parents and it was found that due to trangressive segregation the line no. 40 was significantly superior to the check (DBW-14) and both the parents. Some of the superior performing lines could be used in breeding programs for the trait of interest to breeders. In addition, the DH lines that showed significantly higher yield than the parents in the present experiment, can be used as a parent in breeding for heat tolerance.

CONCLUSION:

Through the present study we evaluated genotype and environmental performance of double haploid lines of wheat over a wide range environments. Stability analysis demonstrated the highly responsive lines from 120 double haploid population in changing environments in terms of enhanced yield under heat stress condition. All the stable lines exhibited 5-20% higher yield over the parent. In this study 3 lines showed stability for GFD, 5 lines for GY whereas 13 lines were found stable for CT. Line nos. 22 and 106 were found to be stable in terms GY and CT when considering all the three parameters of stability for four environments studied. The negative correlation of CT with GY indicates this trait to be taken as a selection parameter. In addition, the DH lines, that showed significantly higher yield than the parents under heat stressed condition, can be used as a parent in breeding for heat tolerance. This study involved a limited number of DH lines but still delivered significantly superior lines for heat tolerance. This suggests that with an increase in population size greater opportunities for obtaining improvements in heat stress tolerance would be available.

Table :1 Mean sum of squares for grain yield ,grain filling duration, canopy temperature in wheat doubled haploid population derived from the cross Sokol/Krichauff under heat stress (late sown) condition in four years of testing in EGP of India

		Grain yield	Grain Filling Duration	Canopy temperature
Rep within Env.	8.00	39517.09	3.19	1.04**
Varieties	119.00	451049.16	7.11	0.68**
Env.+ (Var.* Env.)	360.00	876119.94**	22.86**	0.62**
Environments	3.00	67930848.00**	2144.60**	37.77**
Var.* Env.	357.00	312634.78	5.03	0.31
Environments (Lin.)	1.00	203792560.00**	6433.80**	113.32**
Var.* Env. (Lin.)	119.00	154208.92	2.82	0.34
Pooled Deviation	240.00	388582.31**	6.09**	0.29**
Pooled Error	952.00	9327.38	0.72	0.21
Total	479.00	770517.81	18.95	0.64
Heritability in percentage (%)		**65**	**55**	**40**

Table -2 Range of wheat DH line and their parental genotype Sokoll/Krichauff for grain yield (GY), grain filling duration(GFD) and canopy temperature (CT) in four years (2007-2011) of testing under late sown (heat stressed) conditions in EGP of India

	Grain yield (Kg/ha)				**GFD** (days)				**CT** (°C)			
Genotype/Year	**2007-08**	**2008-09**	**2009-10**	**2010-11**	**2007-08**	**2008-09**	**2009-10**	**2010-11**	**2007-08**	**2008-09**	**2009-10**	**2010-11**
Sokoll	2052.1±62.11	2903.2.±309.92	2505.2±128.17	2164±40.18	26±0.59	31±1.33	25±0.88	26±0.40	29.8±0.88	29±0.33	29.9±0.33	30.9±0.33
Krichauff	2637.45±124.69	3220.56±238.34	2151.85±88.09	2692.50±59.62	30±0.33	31±0.66	30±0.33	29±0.88	29.5±0.33	29±0.33	29.4±0.66	30.9±0.33
DH Range	1000-3528.01	1632.41-3958.00	1314.81-3845.00	1281.25-3905.00	20-31	27-30	26-31	21-31	29-29.2	28-31.7	29.4-30.9	30.1-31.5

±Indicate standard error

Table:3 Correlation among yield and yield traits across four years (2007-2011) of testing of 120 DH lines of the corss Sokol/Krichauff in eastern Gangetic plains of India

	GFD	**GY**	**CT**
GFD	1	0.319**	0.264**
GY	0.319**	1	-0.149
CT	0.264**	0.149	1

**. Correlation is significant at the 0.01 level (2-tailed).

Table 4. Mean performance of best 20 DH lines out of total population of 120 for grain yield with respect to their grain filling duration (GFD) and canopy temperature (CT) in four years (2007–11) of testing at Varanasi, India

Original Line No.	Pedigree name	GY (Kg/ha)	GFD (Days)	CT (°C)
40	Sokoll / Krichauff-DH -40	3809.26	30	30.2
41	Sokoll / Krichauff-DH -41	2983.51	30	30.8
12	Sokoll / Krichauff-DH -12	2957.58	30	30.1
35	Sokoll / Krichauff-DH -35	2881.60	31	30.0
74	Sokoll / Krichauff-DH -74	2854.34	27	29.9
77	Sokoll / Krichauff-DH -77	2792.47	28	29.5
8	Sokoll / Krichauff-DH -8	2775.06	28	30.3
75	Sokoll / Krichauff-DH -75	2772.34	27	29.9
5	Sokoll / Krichauff-DH -5	2731.04	28	29.2
42	Sokoll / Krichauff-DH -42	2725.98	30	30.5
7	Sokoll / Krichauff-DH -7	2710.88	29	30.4
46	Sokoll / Krichauff-DH -46	2709.84	29	30.0
28	Sokoll / Krichauff-DH -28	2709.12	29	30.8
3	Sokoll / Krichauff-DH -3	2672.76	26	29.6
54	Sokoll / Krichauff-DH -54	2660.36	31	31
106	Sokoll / Krichauff-DH -106	2655.73	29	30.2
30	Sokoll / Krichauff-DH -30	2646.06	29	29.7
95	Sokoll / Krichauff-DH -95	2625.00	27	29.4
70	Sokoll / Krichauff-DH -70	2615.34	31	30.1
15	Sokoll / Krichauff-DH -15	2614.18	29	30.6
151(P1)	Sokoll	2406.13	27	29.9
154(P2)	Krichauff	2675.59	30	29.7
Check	DBW-14	3570.00	30	29.1
	LSD (1%)	208.95	0.82	0.18

Table.5 Late sown data of the Sokoll/Krichauff DH Population for the GY, GFD and CT

Line No.	Pedgree name	GY (kg/ha)	Line No.	Pedgree name	GFD (days)	Line No.	Pedgree name	CT (°C)
40	Sokoll /Krichauff-DH -40	3809.25	70	Sokoll / Krichauff-DH -70	31	144	Sokoll / Krichauff-DH- 144	29.1
41	Sokoll / Krichauff-DH -41	2983.50	27	Sokoll / Krichauff-DH -27	31	105	Sokoll / Krichauff-DH -105	29.1
12	Sokoll / Krichauff-DH -12	2957.58	54	Sokoll / Krichauff-DH -54	31	5	Sokoll / Krichauff-DH -5	29.2
35	Sokoll / Krichauff-DH -35	2881.59	14	Sokoll / Krichauff-DH -14	31	143	Sokoll / Krichauff-DH -143	29.2
74	Sokoll / Krichauff-DH -74	2854.34	106	Sokoll / Krichauff-DH -106	31	156	Westonia	29.3
77	Sokoll / Krichauff-DH -77	2792.46	35	Sokoll / Krichauff-DH -35	31	14	Sokoll / Krichauff-DH -14	29.3
8	Sokoll / Krichauff-DH -8	2775.05	66	Sokoll / Krichauff-DH -66	31	113	Sokoll / Krichauff-DH -113	29.3
75	Sokoll / Krichauff-DH -75	2772.33	41	Sokoll / Krichauff-DH -41	30	153	SAMNYT 19	29.3
5	Sokoll / Krichauff-DH -5	2731.04	11	Sokoll / Krichauff-DH -11	30	147	Sokoll / Krichauff-DH -147	29.4
42	Sokoll / Krichauff-DH -42	2725.98	12	Sokoll / Krichauff-DH -12	30	13	Sokoll / Krichauff-DH -13	29.4
7	Sokoll / Krichauff-DH -7	2710.88	64	Sokoll / Krichauff-DH -64	30	95	Sokoll / Krichauff-DH -95	29.4
46	Sokoll / Krichauff-DH -46	2709.83	142	Sokoll / Krichauff-DH -142	30	4	Sokoll / Krichauff-DH -4	29.4
28	Sokoll / Krichauff-DH -28	2709.123	102	Sokoll / Krichauff-DH -102	30	61	Sokoll / Krichauff-DH -61	29.5

28	Sokoll / Krichauff-DH -28	2709.123	102	Sokoll / Krichauff-DH -102	30	61	Sokoll / Krichauff-DH -61	29.5
3	Sokoll / Krichauff-DH -3	2672.76	40	Sokoll / Krichauff-DH -40	30	92	Sokoll / Krichauff-DH -92	29.5
54	Sokoll / Krichauff-DH -54	2660.35	42	Sokoll / Krichauff-DH -42	30	132	Sokoll / Krichauff-DH -132	29.5
106	Sokoll / Krichauff-DH -106	2655.72	138	Sokoll / Krichauff-DH -138	30	142	Sokoll / Krichauff-DH -142	29.5
30	Sokoll / Krichauff-DH -30	2646.06	21	Sokoll / Krichauff-DH -21	30	141	Sokoll / Krichauff-DH -141	29.5
95	Sokoll / Krichauff-DH -95	2625.00	16	Sokoll / Krichauff-DH -16	30	130	Sokoll / Krichauff-DH -130	29.5
70	Sokoll / Krichauff-DH -70	2615.33	52	Sokoll / Krichauff-DH -52	30	77	Sokoll / Krichauff-DH -77	29.5
15	Sokoll / Krichauff-DH -15	2614.17	36	Sokoll / Krichauff-DH -36	30	96	Sokoll / Krichauff-DH -96	29.5
151 (P1)	Sokoll	2406.13	151	Sokoll	27	151	Sokoll	29.9
154 (P2)	Krichauff	2675.58	154	Krichauff	30	154	Krichauff	29.7
Check	DBW-14	3570.00		DBW-14	30		DBW-14	29.1
	LSD (1%)	208.95		LSD	0.82		LSD	0.18

Table 6: Estimation of stability parameters for heat tolerant traits in Doubled haploid wheat.

Line no.	GFD	Mean	bi	S^2di
	Pedgree			
39	Sokoll / Krichauff-DH -39	28.83	0.702	0.778
63	Sokoll / Krichauff-DH -63	28.25	0.791	-0.367
69	Sokoll / Krichauff-DH -69	28.33	0.778	0.149
151	Sokoll	28.25	1.196	9.04
154	Krichauff	28.58	1.062	1.39
	Yield			
	Pedgree			
3	Sokoll / Krichauff-DH -3	2672.76	1.28	5088.75
5	Sokoll / Krichauff-DH -5	2731.03	1.18	-3908.08
22	Sokoll / Krichauff-DH -22	2613.19	1.63	9812.24
35	Sokoll / Krichauff-DH -35	2881.59	08972	13423.42
106	Sokoll / Krichauff-DH -106	2655.72	1.34	4314.53
151	Sokoll	2406.13	0.925	2075.77
154	Krichauff	2675.58	0.999	-1667.44
	CT			
	Pedgree			
10	Sokoll / Krichauff-DH -10	29.91	0.731	0.021
11	Sokoll / Krichauff-DH -11	30.16	1.35	0.423
15	Sokoll / Krichauff-DH -15	30.66	1.55	-0.135
16	Sokoll / Krichauff-DH -16	30.41	0.704	-0.183
18	Sokoll / Krichauff-DH -18	30.66	1.26	0.183
20	Sokoll / Krichauff-DH -20	30.58	1.69	-0.079
21	Sokoll / Krichauff-DH -21	30.25	1.27	-0.158
22	Sokoll / Krichauff-DH -22	29.91	1.03	0.329
23	Sokoll / Krichauff-DH -23	29.83	1.53	-0.151
24	Sokoll / Krichauff-DH -24	30.66	0.961	-0.093
63	Sokoll / Krichauff-DH -63	29.75	2.13	0.112
69	Sokoll / Krichauff-DH -69	30	1.08	0.123
106	Sokoll / Krichauff-DH -106	30.25	1.516	-0.034
151	Sokoll	29.91	0.85	-0.070
154	Krichauff	29.75	0.94	0.072

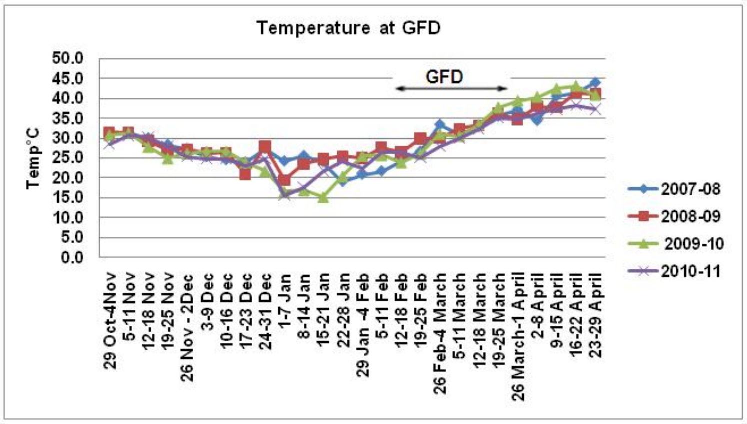


Fig.1. Daily mean temperatures of wheat growing seasons (2007-08, 2008-09 , 2009-10 and 2010-11)

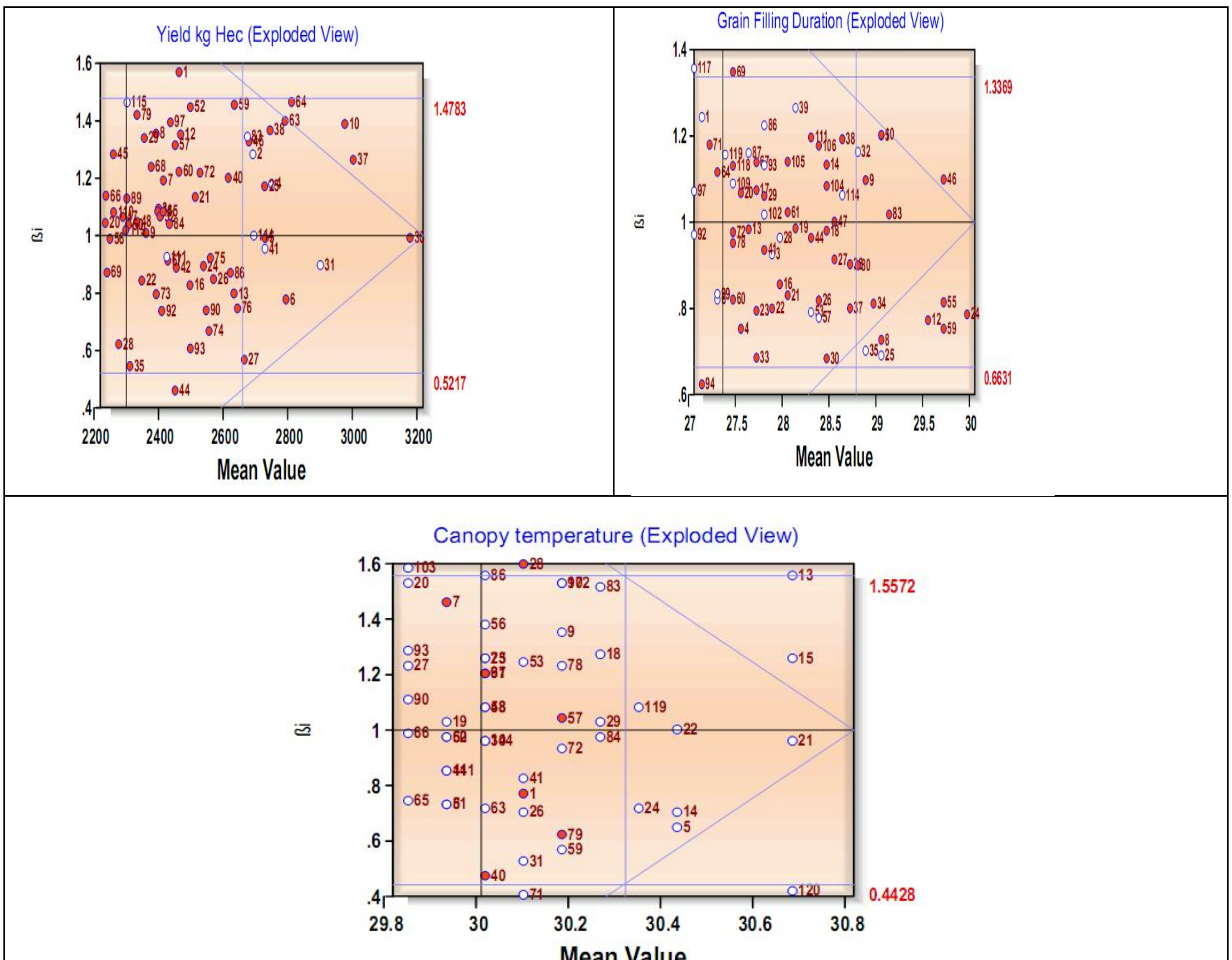


Figure 2: Graphical representation of stable DH lines for the traits grain yield ,grain filling duration, canopy temperature in heat stress condition

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