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Characterization of RILs for Heat Tolerance for Physio-morphological Trait in Bread Wheat (*Triticum aestivum* L.)

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ABSTRACT: Wheat (*Triticum aestivum* L.) as a major cereal grass of *Graminae* family and genus *Triticum*, and is the world's largest grown cereal crop. Among all abiolic stresses high temperature is an major abiotic stress limiting production of crops in all regions were wheat is grown. Climate change is reported to loss of agricultural production the most, with severe effects of high temperature, challenging researchers toward developing strategies to counter stress. These constraints to global food supply encourage research for development of heat resistant genotypes, which are resilient to climate change. In this research we use RIL population for studying heat stress and in understanding the physiological and morphological mechanism of heat tolerance. In current research a set of 30 bread wheat recombinant inbred lines which were derived from three cross *viz.* LOK-1 and HUW- 468, LOK 1 and HUW 234 and Raj-4014 and PBN-51, along with two controls namely PBN-51 and HUW-234, were studied using a RBD with three replications grown in *Rabi* 2018-19. Analysis of variation exhibited significant variation in mean sum square for genotypes in all the charcters under study, which showed the existence of genetic variation among the RIL population developed from this cross. Grain yield per plant and tillers per plant showed to moderate heritability with low PCVand GCV.

Keywords: RIL, Heat tolerance, Physiological, morphological, genetic variation, PCV, GCV.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is the major food crop for majority of world population. It is a hexaploid (AABBDD) having chromosome number of 42 (2n=6x=42) with genome size of 16 GB. Wheat is the major staple food crop and a main source of energy for world population. Wheat production, at global level is about 778.6 million metric tonnes, with area under cultivation of 223.40 million hectares and productivity of 3.49 metric tonnes per hectare. In India it is major crop with 31.62 million hectare area under cultivation, producing about 109.52 million metric tonnes with productivity of 3.46 metric tonnes per hectare.

Effect of biotic stress are major field to study for the crop scientists and plant breeders so as to deduce better approach to prevent losses caused by these stresses. High temperature at anthesis stage results in yield loss because of the narrow optimum temperature range of fertilization as high temperature disrupts fertilization via the abnormal development of reproductive organs, such as the ovule or pollen tube, which in turn increases grain abortion (Prasad and Djanaguiraman, 2014). High temperature increases the fluidity of cell and organelle lipid membranes, interfering with the ability of membranes to regulate passage of metabolites in and out of the cell (Niu and Xiang, 2018). Under a high emission scenario, global 80 mean temperature will continue to rise by at least 4°C towards the end of this century 81 (Pachauri *et al.*, 2014). High temperature is major stress decreasing crop and cereal production in all regions of the major wheat growing areas (Brenchley *et al.*, 2012). High temperatures can severely effect developmental and growth processes in wheat. Morever, heat stress alters agronomic traits critical stages of wheat growth, but pre-flowering and anthesis stages are mostly affected as they are sensitive to high temperature compared to post-flowering stages of wheat (Shah *et al.*, 2003). High temperature stress in the recent years has been major factor affecting the wheat productivity especially, in the arid and semi arid regions of the world. The earth temperature is constantly increasing. The cultivation of wheat is limited by temperature at both ends of the cropping season (Bhanu *et al*., 2018).

Heat stress effects tiller production, biomass production, grains per spike and grain size at crucial stage of wheat growth. Heat stress in later stages of crop growth leads to decrease in number of kernels/ear and kernel weight which finally leads to decrease in yield (Gupta *et al.,* 2001).

Grain yield is a complex trait and highly influenced by many physiological and morpholoogical characters. As direct selection for yield as such could be baised so we need a selection criteria which depends upon the information on the genetic variability and association of morpho-agronomic character with grain yield (Ali *et al*., 2008). Information about the genetic variability, among genotypes and inheritance of the genetic characters is an important task in genetic improvement of any crop plant. Knowledge about heritability helps the plant breeder in estimating the behaviour of the succeeding generation and for selections of desired genotypes. Genetic advance is a significant selection parameter that assists breeder in a selection program. It indicates the extent of the expected genetic gain from one cycle of selection (Naik *et al.,* 2015). Heritability estimates with genetic advance is normally more powerful than heritability estimates alone in forecasting the genetic gain under selection (Kumar *et al.,* 2015).

Success of breeding crops for abiotic stress mainly depend on study of physiological characters of plants, which should be considered as the main contributing parameters for heat tolerance in advanced generation. These genetic parameter could be used in selecting the genotypes which are tolerance to high temperature. Various selection basis for genetic parameters were studied to envisage the morpho-physiological trait under selection for the development of yield resistant cultivar in wheat. Although many tolerant cultivar have been identified earlier in wheat germplasm (Reynolds *et al*., 2001), the characterization of recombinant inbred lines for heat tolerance was undertaken with objectives to characterize elite Rils for morpho-physiological traits related to heat and productivity.

MATERIAL AND METHODS

A. Plant material and field trails

This current experiment was carried out at the Research Area of Division of Plant Breeding and Genetics, Sher-e-Kashmir University of Agricultural Science and Technology of Jammu, Chatha, which is located at an altitude of 332 m above sea level with 320 39$''$ degree N latitude and 740 degree 58$''$ E longitude and sub-tropical area.

The experimental material composed of 450 RILs population derived from three crosses (LOK-1 × HUW- 468, LOK 1 × HUW 234 and Raj-4014 × PBN-51). These were sown during *Rabi* 2017-2018 in an augmented design with two contro; variety, PBN 51 (heat tolerant) and HUW 234 (heat susceptible) line after each 20 lines. Further these 450 RILs were studied for thirteen morpho-physiological traits. Ten RILs from each of the individual three crosses were selected based on mean data of 450 RILs showing better response to heat stress. Trait data from 30 RILs along with two checks, PBN 51 and HUW 234 were studied during *Rabi* 2018-2019. Two rows of five meter using experimental design of RBD design in three replication for better understanding of variability in 30 RILs for thirteen morphological and physiological traits to select best performing RILs for heat tolerance and grain yield was done.

The meteorological data during stage of heat stress was obtained from Meteorological Station, SKUAST Chatha Jammu. The maximum temperature ranged from 43.1^{o}C in fourth week of May to 15^{o}C in first week of November, while minimum temperature recorded ranged from 1.7^{o}C in last week of December to 21.7^{o}C in first week of May. The experimental crop received highest maximum rainfall of 67.8 mm in third week of February and minimum rainfall of .6 mm in second week of May. 77 days recorded temperature above 25^{o}C. Days during reproductive stages coincided with flowering and grain filling stages which are most crucial stages for yield determination. The recorded data was subjected to analysis using Windostat.

RESULT AND DISCUSSION

A. Analysis of variance and components of variation

Measure of variability indicated mean square differences among thirty RILs were shown to be significant for most of characters. Average performance of thirty RILs and two checks for physio-morphological characters is represented in Table 1. Analysis of Variance (Table 2) for 13 morphological and physiological traits as a measure of variability indicated mean square for difference in RILs were highly significant for all the characters. The present findings corroborated the earlier reports of (Karagoz *et al.*, 2005) for of days to flowering and height of plant, for relative water content by (Merah, 2001), for number of grain per spike, thousand grain weight and grain yield per plant by (Ali *et al.*, 2008), for canopy temperature by (Saint *et al.*, 2010), for flag leaf area by (Khaliq *et al.*, 2008) for RWL by (Kumar *et al.*, 2007). In our investigation relative water loss was lower at high temperature in PBN-51 followed by RIL 14 and 12 of cross 2 showing low water losses at high temperature. The high RWC and low RWL have been suggested as important indicators of water status (Farshadfar *et al.*, 2011). (Moradi *et al.*, 2015) also obtained similar results for physiological traits such as relative water content and chlorophyll content. Kutlu and Kinaci (2010) also abtained similar result for grin yield and plant height. Futher MSS due to genotypes showed significant values for all the character studied, suggesting that high genetic variability is present among the RILs for all the thirteen traits. Large variability among RILs represents presence of wide diversity among RILs. Effect of environment was less on almost all the thirteen characters. This was represented by small gap between phenotypic and genotypic coefficient of variation. Moderate values of GCV and PCV were reported for canopy temperature, plant height, flag leaf area and thousand grain weight.

Table 1: Mean performance of 32 wheat genotype for various physiological and morphological characters.

Sr. No.	GENOTYPE	DTF	DTM	PH (cm)	TPP	G/E	TGW (g)	FLA (cm^2)	CC (SPAD Unit)	RWC (%)	RWL (%)	CT (^{0}C)	CTD (^{0}C)	GY^{-1} (g)
1.	CROSS 1 RIL 1	97.59	119.00	96.78	10.35	42.66	37.82	34.45	38.57	63.72	77.58	20.70	8.24	16.20
2.	CROSS 1 RIL 2	89.66	126.66	93.43	9.51	39.44	36.81	33.67	37.86	57.62	77.26	21.07	7.64	15.12
3.	CROSS 1 RIL 3	93.66	122.33	85.58	9.33	42.33	32.77	33.38	40.00	58.28	78.48	26.15	7.66	16.81
4.	CROSS 1 RIL 4	94.66	121.66	99.12	8.77	41,77	33.24	33.84	37.21	63.50	77.99	19.44	7.7	15.99
5.	CROSS 1 RIL 5	96.66	124.66	81.27	9.22	40,44	28.72	28.36	37.72	60.86	75.21	20.70	7.92	15.47
6.	CROSS 1 RIL 6	97.66	125.66	98.95	9.44	43.89	30.66	30.91	36.94	52.10	74.92	24.85	7.91	14.41
7.	CROSS 1 RIL 7	99.66	111.66	85.01	8.56	42.67	29.80	29.98	38.39	58.24	74.13	21.08	7.72	15.64
8.	CROSS 1 RIL 8	97.16	109.00	80.03	8.44	39.11	28.14	31.30	37.16	50.78	74.69	20.72	7.21	17.75
9.	CROSS 1 RIL 9	104.66	111.33	82.06	9.00	40.66	27.14	22.29	37.76	56.02	73.00	27.29	5.92	17.35
10.	CROSS 1 RIL 10	100.66	108.66	86.46	9.88	45.89	27.27	30.01	38.49	53.04	74.72	18.62	7.78	15.91
11.	CROSS 2 RIL 11	104.66	123.00	78.99	11.46	41.33	25.82	34.73	39.33	56.59	76.58	24.39	7.66	16.21
12.	CROSS 2 RIL 12	98.33	130.87	86.52	9..54	39.22	22.39	35.23	40.3	55.67	72.92	20.59	8.23	17.59
13.	CROSS 2 RIL 13	106.00	125.66	88.69	9.78	42.11	24.23	34.18	38.00	58.71	73.16	22.43	7.65	17.57
14.	CROSS 2 RIL 14	94.66	127.33	85.98	9.33	40.00	28.71	29.08	40.02	60.42	72.13	24.64	7.86	15.78
15.	CROSS 2 RIL 15	103.66	122.33	69.42	9.22	39.78	29.34	31.15	41.44	53.18	76.40	21.02	7.30	16.42
16.	CROSS 2 RIL 16	93.16	119.33	80.25	8.89	40.22	27.63	33.76	39.7	60.95	72.46	23.23	7.20	14.46
17.	CROSS 2 RIL1 7	96.66	127.33	77.09	8.78	43.22	29.78	32.58	38.37	58.40	77.26	18.28	7.56	16.12
18.	CROSS 2 RIL 18	95.66	125.66	86.6	8.89	41.00	30.11	31.10	39.82	50.33	73.67	20.91	7.50	15.40
19.	CROSS 2 RIL 19	101.66	114.00	87.76	8.22	45.89	30.56	32.75	38.66	54.49	80.86	20.14	6.97	15.17
20.	CROSS 2 RIL 20	89.66	123.33	80.13	7.44	38.55	31.15	35.23	39.76	59.40	78.23	21.29	7.68	17.73
21.	CROSS 3 RIL 21	104.66	113.66	81.75	8.00	41.77	31.99	32.72	39.36	54.74	76.62	24.14	7.94	16.27
22.	CROSS 3 RIL 22	97.66	113.00	92.49	8.67	38.66	32.15	32.41	37.44	56.78	77.37	17.87	7.89	16.93
23.	CROSS 3 RIL 23	96.12	109.66	87.41	8.67	40,22	38.06	31.95	40.52	59.02	75.80	26.27	7.11	14.82
24.	CROSS 3 RIL 24	95.13	101.00	86.04	8.89	36.11	32.98	21.64	40.09	54.60	72.86	17.74	7.44	16.50
25.	CROSS 3 RIL 25	103.66	105.33	71.44	8.56	41.89	36.71	31.72	38.87	54.48	74.10	26.86	7.27	16.76
26.	CROSS 3 RIL 26	99.66	119.66	62.81	7.89	41.22	29.95	30.37	37.22	53.52	76.36	21.47	7.93	15.46
27.	CROSS 3 RIL 27	92.66	121.00	73.94	8.56	41.22	31.99	36.10	37.75	57.79	77.09	17.42	7.16	17.65
28.	CROSS 3 RIL 28	99,66	113.66	79.11	6.67	38.89	30.83	22.16	37.1	56.98	75.63	18.01	8.08	16.53
29.	CROSS 3 RIL 29	108.66	128.46	84.5	9.00	39.77	31.55	32.28	38.43	63.11	75.34	25.19	7.69	15.32
30.	CROSS 3 RIL 30	78.41	126.84	76.18	8.33	37.77	33.29	25.32	38.47	50.88	74.74	21.51	7.14	18.07
31.	Check1 HUW234	101.66	114.00	108.42	10.33	34.15	27.74	36.50	39.42	60.78	76.45	16.83	8.25	19.41
32.	Check 2 PBN 51	102.00	117.66	116.78	10.00	37.08	33.81	31.42	41.78	59.59	67.52	20.64	6.22	20.15
	MEAN	**98.29**	**118,85**	**85.37**	**9.28**	**40.59**	**30.72**	**31.51**	**38.85**	**57.13**	**75.36**	**21.61**	**7.54**	**16.47**
	C.V(%)	**2.41**	**5.98**	**9.65**	**11.26**	**5.80**	**11.95**	**6.13**	**4.27**	**3.92**	**4.12**	**12.87**	**8.14**	**6.17**

PH-Plant height (cm), G/E-Number of grain per ear, TPP- Number of tillers per plant, FLA-Flag leaf area (cm^2), DTM- Number of days to maturity, DTF- Number of days to flowering , TGW-Thousand grain weight (g), CT=Canopy temperature (^{0}C), CTD-Canopy temperature depression (°C), RWC-relative water content (%), CC-Chlorophyll Content (SPAD unit) , RWL-Relative water loss (%), GY^{-1}Grain yield per plant (g)

In their study, Ali *et al.* (2008) reported similar result of GCV and PCV for grain yield per plant and thousand grain weight. Similar result was also shown by **(**Aharizad *et al*., 2012). **(**Ashinie *et al.*, 2011) also obtained similar result for plant height, relative water content and grain yield. Low values of GCV and PCV was seen for tillers per plant, number of days to maturity, number of grains per ear, grain yield per plant, chlorophyll content, relative leaf water content, canopy temperature depression, and relative water loss. These results are in conformation to the report of (Maurya *et al*., 2014). Variability is the only heritable from one generation to the other generations. It has to be considered in conjugation with the genetic advance for accurate results. Flag leaf was reported to show high heritability with high genetic advance as per cent of mean, grain yield per plant exhibited medium heritability with moderate genetic advance, relative water content exhibited moderate genetic advance and high heritability. Akanda *et al.*, (1997) reported that high genotypic coefficient of variation along with high heritability and genetic advance predicts more accurate information than other parameters. Canopy temperature which exhibited genetic advance with moderate heritability were same as obtained by (Riaz-ud-din *et al*., 2010) for flag leaf area, (Kumar *et al*., 2014) for grain yield per plant, (Rathwa *et al*., 2018) for canopy temperature. (Cheema *et al.*, 2006) also obtained similar result for thousand grain weight and yield signifying efficiency of selection for the improvement of these characters for future breeding programmes.

Table 2 : Mean sum of squares obtained from ANNOVA for thirteen traits in thirty two wheat genotype.

		Mean sum of square							
Source of variation	**d.f**	**Number of days to fifty per cent flowering**	**Number of day to maturity**	**Plant Height (cm)**	**Number of tillers per plant**	**Number of grains per ear**	**Thousand Grain weight (g)**	**Flag leaf area (cm^2)**	**Grain yield [1] (g)**
Replication	2	148.88	1130.04	12223.32	0.002	153.18	35.41	263.87	1.95
Genotype	31	108.08**	171.61**	346.89**	2.25**	18.74**	39.99**	45.48**	5.23**
Error	62	5.62	5.62	67.83	0.73	5.52	13.48	3.73	1.03

*,** significantly at 5% and 1% levels, respectively

Table 3: Mean, range, phenotypic and genotypic coefficient of variation, heritability and genetic advance for various traits in thirty two wheat genotype under study.

Character	Mean	Range	Environmental coefficient of variance (%)	Genotypic coefficient of variance (%)	Phenotypic coefficient of variance (%)	h² (Broad sense heritability)	Genetic advance	Expected genetic advance as % of Mean
Number of days to fifty per cent flowering	98.29	78.41-108.66	2.41	5.94	6.42	85.9	11.15	11.34
Number of days to maturity	118.85	101.00-130.87	5.98	5.34	8.02	44.4	8.71	7.33
Plant height (cm)	85.57	62.81-116.78	9.62	11.27	14.82	57.8	15.10	17.66
Number of tillers per plant	09.28	6.67-11.46	9.54	7.92	12.41	40.8	0.93	10.42
Number of grains per ear	40.59	34.15-45.89	5.80	5.16	7.76	44.3	2.87	7.08
Thousand Grain weight (g)	30.72	22.39-38.06	11.91	9.67	15.37	39.6	3.853	12.54
Flag leaf area (cm^2)	31.51	21.64-36.50	6.13	11.83	13.33	78.8	6.82	21.65
Chlorophyll content (SPAD Unit)	38.85	36.94-41.78	4.21	2.23	4.77	21.9	0.83	2.15
Relative water content (%)	57.13	50.13-63.72	2.94	6.23	6.89	81.7	6.61	11.60
Relative water loss (%)	75.36	67.52-80.86	4.12	2.27	4.72	23.3	1.70	2.26
Canopy temperature (°C)	21.61	16.83-27.29	12.87	11.37	17.18	43.8	3.35	15.50
Canopy temperature de pression (°C)	07.54	5.92-8.24	8.17	4.95	9.56	26.9	0.39	5.29
Grain yield^{-1} (g)	16.47	14.46-20.15	6.17	7.18	9.47	57.5	1.84	11.21

CONCLUSION

Study of GCV and PCV values indicates that characters such as canopy temperature, plant height, flag leaf area and thousand grain weight with high and medium GCV indicating that selection may be effectively based on these characters. Grain yield, tiller per plant and canopy temperature exhibited medium values of heritability with moderate value of genetic advance whereas RWC shows high value of heritability with moderate genetic advance. High value of heritability and high value of genetic gain was reported for flag leaf area. Chlorophyll content showed high value for PBN-51 followed by cross 2 RIL 15, Cross 3 RIL 23 and Cross 3 RIL 24, with nearly same values showing low chloroplast damage due to high temperature.

For relative water content Cross 1RIL 2 showed high value followed by Cross 1 RIL 4, Cross 3 RIL 29 showing better water status under dehydration Some important result can be established from the present study that tillers per plant, chlorophyll content, number of tillers per plant, flag leaf area, canopy temperature and canopy temperature depression are principal selection tools for breeding of high yield varieties of wheat under heat stress.

FUTURE SCOPE

Future breeding programme should focus on developing new varieties that are more adaptive to thermal stress acclimating respiration and photosynthesis in heat stress to avoid the yield losses. Phenotyping is a prerequisite for any successful breeding methodology. In order to successfully develop suich varieties equipped for high temperature acclimation we need to identify the extent of genetic variation in breeding population for various physiological and morphological traits that exists for these traits. Better heat resistant varieties could also be developed by delaying flowering mechanisms so that plant can escape heat stress during reproductive stages. Further we can identify the biochemical mechanisms that confer chloroplast and mitochondrial heat tolerance and use them as a option for developing potential heat resistant cultivars.

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Conflict of Interest. None.

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