

## Study of diversity in thermo-tolerant advanced lines of Wheat (*Triticum aestivum* L.) for Morpho-physiological, Yield and its Contributing Traits

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### Abstract

The present study was carried out to examine the genetic diversity among 151 thermo-tolerant advanced breeding lines of wheat for two experimental years during Rabi 2017-18 and 2018-19. The data were recorded in both the seasons for total 23 traits i.e. 18 quantitative characters to study variability, heritability, genetic advance and genetic divergence and 5 morphological characters. ANOVA showed significant differences for all the characters except flag leaf width, in both experimental seasons and for gluten content in season Rabi 2018-19 indicating the presence of a substantial amount of genetic variability which thus revealed different genetic backgrounds of genotypes. High estimates of both GCV and PCV were observed for biological yield in both experimental years. High heritability with high genetic advance was recorded for all quantitative characters except flag leaf width and gluten content indicating the predominance of additive gene effects and hence the possibilities of effective selection for the improvement of these traits. D<sup>2</sup> analysis grouped these lines into 10 clusters in which cluster VII was biggest cluster with 26 lines. Further diversity analysis revealed that cluster IV and VII followed by Cluster VI and Cluster VII were most divergent from each other, and the genotypes present in them may be used as parents for further hybridization program to develop desirable heat type in wheat. Analysis of 5 morphological characters i.e. leaf type, rachis colour, leaf shape, waxiness and stay green habit, depicted that HUW-213 x NW-1076 followed by HALNA x RAJ-4037 were found above average yielders and terminal heat tolerant primarily due to their semi-erect leaf type, green rachis color, broad leaf shape, stay green habit and less days to maturity as compared to checks.

**Key words:** D<sup>2</sup> analysis, Genetic diversity, Genetic variability, Heat stress, Terminal heat tolerance, *Triticum aestivum* L, Wheat.

## 1. Introduction

Wheat (*Triticum aestivum* L.) is a cereal grass belongs to Gramineae family and having genus *triticum*, wheat is described as the “King of Cereals”. Being the world’s

most widely cultivated cereal crop, it plays an important role in food and nutritional security. It is the second most important cereal crop after rice contributing nearly



35% to the national food basket. Heat stress is a major abiotic factor that substantially reduces wheat grain yield globally, especially in regions that are associated with higher temperature. Generally, wheat is likely to be sown early in October to November to attain maximum period for growth and development toward maturity before the (possible) heat stress. Rice-wheat cropping system results in delay in wheat sowing (Mitra and Patra, 2019) and leads to low crop yield due to the sub-optimal temperature during germination and reproductive growth (Sattar *et al.*, 2010). The most heat-stressed locations of South Asia are the Eastern Gangetic Plains, Central and Peninsular India and Bangladesh. Heat stress is considered moderate in northwestern parts of the Indian Gangetic Plains (Joshi *et al.*, 2007; Singh *et al.*, 2007).

Heat stress or high temperature during crop growing period restricts wheat production and productivity, particularly at germination, anthesis and grain filling stage. A rise in temperature at the time of grain filling is responsible for decline in wheat production in many wheat environments due to heat stress around the world (Rajaram *et al.* 1995). Heat stress for a period of time is sufficient to cause irreversible damage to plant growth and development (Wahid *et al.*, 2017). In wheat, the rate of grain filling decrease due to high temperatures (30 °C) after anthesis (Stone and Nicolas, 1995), while high temperatures before anthesis reduces grain yield. Yields are reduced 3–4 % per 1°C rise above the optimum temperature (15–20 °C) during grain filling (Wardlaw *et al.*, 1989). Wardlaw and Wrigley (1994) observed that a heatwave (35–37 °C) of 3–4 days modifies grain morphology and reduces grain size. A reduction in grain yield up to 23 % can occur in response to short period (4 days) of exposure to high temperature (35 °C) in wheat (Stone and Nicolas, 1994) and 3-day heat treatment (38 °C from 8 am to 5 pm) reduced individual yield component up to 28.3 % (Mason *et al.*, 2010). Therefore, breeding for high-temperature tolerance in wheat is a major objective globally. The development of wheat germplasm adapted to high temperature stress has been a key strategy for reducing the associated loss in grain yield.

It is necessary to investigate the genetic diversity in wheat germplasm to broaden the genetic variation in future wheat breeding. Estimation of genetic distance

is one of the appropriate tools for parental selection in hybridization programs. Genetic diversity can be accessed from pedigree analysis, morphological traits or using molecular markers (Pejic *et al.*, 2008). Genetic diversity is important among all crops for successful production of hybrids and new cultivators. It may provide new sources of tolerance and resistance against several biotic and abiotic stresses (Skovmand *et al.*, 2002). Presence of diversity in the germplasm acts as insurance against future needs and conditions, thereby contributing to the stability of the farming system at local, national and global levels.

Keeping all the facts in view, the present study was planned to evaluate and characterize advanced terminal heat tolerant wheat lines for variability parameters, to estimate genetic diversity in advanced lines of wheat, to identify diverse parents for developing future terminal heat tolerant genotypes and to find out high yielding wheat genotypes for desirable morpho-physiological, maturity and other characters associated with terminal heat tolerance.

## 2. Materials and Methods

The experiment were carried out at Field Experimentation Center, Department of Genetics and Plant Breeding, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj (Allahabad), Uttar Pradesh during *Rabi* 2017-18 and *Rabi* 2018-19 in randomized block design (RBD) with three replications. The experimental materials for the present study consisted of 151 (149+2 Checks i.e. NW-2036 and K-424) advanced lines (F<sub>7</sub> and F<sub>8</sub> in respective growing seasons) of wheat (*Triticum aestivum* L.), were obtained from the department, The sowing was done under late sown conditions on 20<sup>th</sup> December, 2017 and on 19<sup>th</sup> December, 2018, whereas harvesting was followed on 11<sup>th</sup> April, 2018 and 14<sup>th</sup> April, 2019, respectively in both the experimental seasons. Each line was planted in a single line of two meter length in each replication with 25 cm row to row distance. Irrigation of field was done at regular intervals depending upon the rainfall and recommended package of practice was followed to raise a healthy crop. Data was recorded for 18 quantitative characters including growth characters i.e. days to heading (Days), days to 50% flowering (Days), spike length (cm), numbers of grains per spike, flag leaf length (cm), flag leaf width (cm), plant height (cm), numbers of tillers per plant, days to maturity



(days), biological yield per plant (g), grain yield per plant (g), test weight (g), harvest index (%), grain filling period (days), two physiological characters chlorophyll content (%) and membrane stability index (%) and two quality characters namely gluten content (%) and sedimentation value (ml). The data for growth characters were recorded from five randomly selected plants from each genotype in every replication. Data for physiological characters i.e. chlorophyll content was done using SPAD-502 chlorophyll meter, membrane stability index and membrane stability test was done on the basis of electrolyte leakage from flag leaves during anthesis. The sample was taken and exposed to heat at 52° C for 15 minutes and electrolyte leakage is measured by electrical conductivity meter. The membrane thermo stability expressed as the relative injury was calculated by using the formula given by Blum (1988) i.e.  $\text{Relative injury (\%)} = 100 \left[ \frac{1 - T_1/T_2}{1 - C_1/C_2} \right]$  \*100, Where, T is treatment, C is control, and 1 and 2 refer to the first and second readings of conductance, i.e., before and after autoclaving. Quality characters, such as gluten content and sedimentation value were taken after harvesting and grinding of wheat to wheat flour. Data was also collected for five morphological characters related to terminal heat tolerance i.e. leaf type, rachis color, leaf shape, waxiness and stay green habit on visual basis and later compared with other characters responsible for terminal heat tolerance and higher grain yield.

The data collected on 18 quantitative characters for both the experimental seasons i.e. *rabi* 2017-18 and *rabi* 2018-19 was analyzed and pooled effect was calculated separately using Genstat 19<sup>th</sup> edition software. The data recorded was subjected to genetic variability, heritability, genetic divergence to find out most diverse parents for future hybridization programme diversity among the different genotypes was measured with the help of D<sup>2</sup> value between and within clusters. Cluster diagram was drawn to show the relationship between different populations.

### 3. Results and Discussion

Analysis of variance showed significant difference among 151 lines for all the characters under the study for both years. The analysis of pooled data indicated that there is ample scope for selection of promising lines for yield improvement. Variability is measured by estimation of

genotypic and phenotypic variation, genotypic coefficient variation (GCV), phenotypic coefficient of variation (PCV), broad sense heritability (H<sup>2</sup>bs), genetic advance and genetic gain for different quantitative traits. These parameters deserve attention in selection for improvement in the concerned traits. Phenotypic variance was higher than genotypic variance for all the characters in all three experimental data tables and early indicating the influence of environmental effect on these characters. Similar findings were reported by Mishra *et al.* (2013) and Singh *et al.* (2012). The minute difference between the values of genotypic and phenotypic variance for all the characters indicates that the variability present among the lines is mainly due to genetic reason with less influence of environment, and hence heritable.

The high GCV is accompanied by a high PCV was observed for biological yield suggests that the genetic variability is present among the wheat genotypes. Therefore, selection will be more effective in isolating superior genotypes. The scope of selection is limited for the traits where PCV and GCV are moderate to low. Heritability is the measure of extent of phenotypic variance caused by the action of genes, has been widely used to access the degree to which character may be transmitted from parents to offspring. Burton (1952) suggested the genetic variation along with heritability estimated would give a better idea about the efficiency of selection. High heritability observed for almost all the characters except moderate in grain per spike and low for gluten content in 2017-18, whereas moderate for chlorophyll content and harvest index in 2018-19 and moderate for harvest index and gluten content in pooled data (Table 1). A high value of heritability indicates that it may be due to a higher contribution of genotypic components and selection would be easy and effective similar results have been reported by Fellahi *et al.* (2013).

High heritability alone is not enough for selection in an advance generation unless accompanied by a substantial amount of genetic advance. Johnson *et al.* (1995) have shown importance of high heritability with high genetic advance for more reliable conclusions. Genetic advance measures the differences between the main genotypic values of the original population from which these were selected.



Table 1. Pooled estimation of components of variance and genetic parameters for 18 quantitative characters.

Character	Vg	Vp	GCV	PCV	h <sup>2</sup> (bs)	GA	GA as 5% of Mean
Days to heading	58.84	59.81	82.53	83.20	98.38	15.7	168.6
Days to 50% flowering	29.79	32.80	58.72	61.62	90.81	10.7	115.3
Plant height	132.55	140.68	123.86	127.61	94.22	23.0	247.7
Tillers per plant	2.40	2.54	16.68	17.14	94.71	3.1	33.4
Flag leaf length	18.11	18.32	45.78	46.05	98.86	8.7	93.8
Flag leaf width	0.10	0.11	3.45	3.54	95.13	0.6	6.9
Spike length	4.54	4.87	22.92	23.74	93.25	4.2	45.6
Membrane stability index	46.16	49.62	73.10	75.79	93.03	13.5	145.2
Chlorophyll content	17.77	21.17	45.36	49.15	83.97	8.0	85.6
Days to maturity	5.32	5.63	24.80	25.53	94.39	4.6	49.6
Grain filling period	24.09	24.40	52.81	53.14	98.74	10.0	108.1
Test weight	38.91	39.81	67.11	67.88	97.75	12.7	136.7
Grain per spike	76.20	114.00	93.91	114.87	66.84	14.7	158.2
Biological yield	1686.50	1703.91	441.82	444.10	98.98	84.2	905.2
Harvest index	1.63	3.29	13.74	19.52	49.54	1.9	19.9
Gluten content	0.62	1.79	8.49	14.40	34.77	1.0	10.3
Sedimentation value	3.79	4.40	20.96	22.56	86.29	3.7	40.1
Grain yield per plant	11.97	12.05	37.22	37.35	99.35	7.1	76.4

Vg: Vp: GCV: PCV: h<sup>2</sup> (bs): GA:

The estimates of genetic advance as 5% of mean high for both years and pooled data for biological yield, while low estimates for flag leaf width for all three experiments with gluten content in 2017-18 and pooled data (Table 1). High heritability accompanied with high genetic advance was found for almost all the characters in all the three experiments which signify the presence of additive gene effects and the possibilities of effective selection. High heritability with moderate genetic advance was found for harvest index in 2017-18 and gluten content in 2018-19 These results indicates the presence of both additive and non additive gene action. This low genetic advance is due to influence of environment and us traits are less significant for selection and improvement of these characters is possible only through careful directional and restricted selection. Whereas character like flag leaf width was having high heritability with low genetic advance, it is

under control of non-additive genes, limits the scope for improvement through selection. Panse (1957) suggested improvement of such characters through individual plant selection.

Genetic divergence is very helpful to plant breeders, the nature and magnitude of genetic divergence in a population are required for the selection of diverse parents and these upon hybridization leads to a wide spectrum of gene recombination for polygenic traits. Based on pooled data of both the seasons 151 lines of wheat were grouped into ten clusters utilizing D<sup>2</sup> values. Cluster VII constitutes of 26 lines, forming the large cluster followed by cluster III constitutes of 25 lines, cluster I constitutes 22 lines, cluster XI constitutes of 17 lines, clusters V and IX both consists 15 lines, cluster IV constitutes 11 lines, cluster II constitutes 9 lines, cluster VI constitutes 8 lines and cluster VIII is with 3 lines.



Table 2. Intra (Diagonal) and inter cluster average distance ( $D^2$ ) for different quantitative characters in wheat for pooled data of both seasons

Cluster No.	I	II	III	IV	V	VI	VII	VIII	IX	X
I	<b>24.44</b> (4.94)	607.62 (24.65)	3682.39 (60.68)	1451.45 (38.09)	1354.67 (36.81)	668.75 (25.86)	3998.84 (63.24)	187.88 (13.71)	1503.83 (38.78)	2060.76 (45.39)
II		<b>21.46</b> (4.63)	3633.32 (60.28)	1377.17 (37.11)	1352.52 (36.78)	612.66 (24.75)	3993.02 (63.19)	140.18 (11.84)	1446.92 (38.04)	2081.35 (45.62)
III			<b>21.04</b> (4.59)	1371.89 (37.04)	1337.07 (36.57)	620.16 (24.90)	3956.04 (62.9)	153.44 (12.39)	1460.16 (38.21)	2069.97 (45.5)
IV				<b>36.77</b> (6.06)	289.86 (17.03)	722.49 (26.88)	4162.2 (64.51)	163.18 (12.77)	1484.75 (38.53)	2285.51 (47.81)
V					<b>20.06</b> (4.48)	625.21 (25.01)	3892.16 (62.39)	162.38 (12.74)	1462.12 (38.24)	2024.94 (44.99)
VI						<b>29.03</b> (5.39)	4088.72 (63.94)	143.02 (11.96)	1576.91 (39.71)	2147.67 (46.34)
VII							<b>21.26</b> (4.61)	155.33 (12.46)	1408.01 (37.52)	2031.08 (45.07)
VIII								<b>28.35</b> (5.32)	1576.9 (39.71)	2277.69 (47.73)
IX									<b>21.36</b> (4.62)	2098.49 (45.81)
X										24.98 (4.99)

The highest intra cluster distance was recorded for cluster IV (36.77) followed by cluster VI (29.03), while the least intra cluster was recorded for cluster V (20.06). The inter cluster  $D^2$  value was maximum between cluster IV and VII (4162.2) followed by cluster VI and VII (4088.7). The intra and inter cluster average distance among 10 clusters were variable. The lowest inter-cluster distance was observed between cluster II and VIII (140.18). The inter-cluster distance in all the clusters were higher than intra-cluster distance indicating wider genetic diversity among genotypes different groups. Clusters IV and VI having highest inter-cluster distance from cluster VII, respectively, therefore, the high yielding genotypes from the cluster IV i.e. K-710 x AAI-12 and cluster VI i.e. Raj- 1480 x Raj-3077 and Raj-4037 x Raj-3777 can be crossed with average yielder genotypes of cluster VII i.e.

NW- 4035 and SAW- 03-02 x Raj-4026, offers us with an opportunity to utilize these lines in a crossing program which may yield transgressive segregates.

For terminal heat tolerance, best suitable morphological characters in wheat are semi-erect leaf type for contribution in photosynthesis, dark green to green rachis colour due to high amount of chlorophyll, broad leaf shape for large area for photosynthesis, presence of waxiness and stay green habit until maturity (Adu *et al.*, 2011). Genotypes HUW-510 x PBW-524 and AAI-12 x K-816 possess all the above mentioned morphological characters responsible for heat tolerance, whereas, HUW-213 x NW-1076, HALNA x RAJ-4037, PHS-06-23 x HUW-510, K-9422 x SAW-336, K-616 x AAI-12, K-710 x AAI-12, AAI-2 x K-8962 and PHS-623 x HUW-510-2 are also having four desirable morphological characters except green rachis colour.





Table 3. Comparison of morpho-physiological, maturity and yield related characters for terminal heat tolerance

Sr. No.	Line	Leaf type	Rachis color	Leaf shape	Waxiness	Stay green habit	Membrane stability index	Chlorophyll content	Days to maturity	Grain filling period	Grain yield per plant
1	HUW-213 x NW-1076	Semi Erect	Green	Broad	Present	Yes	33.05	44.57	119.00	37.00	18.20
2	HALNA x RAJ-4037	Semi Erect	Green	Broad	Present	Yes	38.97	47.51	117.83	35.83	17.30
3	K-9422 x SAW-336	Semi Erect	Green	Broad	Present	Yes	35.63	40.92	121.83	33.33	15.20
4	HALNA x RAJ-1488	Semi Erect	Green	Broad	Absent	Yes	31.90	52.38	124.17	43.17	14.33
5	PHS-06-23 x HUW-510	Semi Erect	Green	Broad	Present	Yes	37.73	42.12	121.17	35.17	13.57
Chk-1	NW-2036	Semi Erect	Green	Medium	Present	No	32.03	49.93	119.50	37.50	15.67
Chk-2	K-424	Droopy	Pale Green	Narrow	Present	Yes	34.44	46.62	121.50	38.00	16.10

Table 3 shows the comparison of morphological and quantitative characters responsible for terminal heat tolerance with higher grain yields and states that HUW-213 x NW-1076 followed by HALNA x RAJ-4037, K-9422 x SAW-336, etc having above average yields primarily due to their semi erect leaf type, green rachis color, broad leaf shape, stay green habit and less days to maturity as compared to checks *viz.*, NW-2036 and K-424 and hence the above genotypes are found tolerant to terminal heat stress suggested by Trethowan and Reynolds (2007).

#### 4. Conclusion

The experimental results suggested that significant variation exists among 151 wheat lines. Based on *per se* performance, high heritability along with high genetic advance as 5% of mean for grain yield per plant, biological yield, grain filling period, days to heading and test weight was observed which revealed that these characters may be used as selection indices for genetic improvement of wheat lines. The less difference between GCV and PCV revealed that there was very low or no influence of environment on the expression of various characters.

In the present study, 151 wheat lines delineated into 10 clusters, clusters IV and VII followed by clusters VI and

VII were more diverse from each other. Therefore, the genotypes from the cluster IV i.e. K-710 x AAI-12 and cluster VI i.e. Raj- 1480 x Raj-3077 and Raj-4037 x Raj-3777 can be crossed with average yielder genotypes of cluster VII i.e. NW- 4035 and SAW- 03-02 x Raj-4026, as clusters IV and VI having highest inter-cluster distance from cluster VII, respectively, It is desirable to use most distant lines present in these clusters for future breeding for higher yields in heat stress environments, as the diversity present in these clusters are supposed to provide a broad spectrum of variability and heterosis for grain yield under terminal heat stress.

A successful attempt was made to record and observe the morphological characters based on terminal heat tolerance. Lines *viz.* HUW-510 x PBW-524 and AAI-12 x K-816 having semi-erect leaf type, dark green rachis colour, broad leaf shape, presence of waxiness and having stay green habit were found to be terminally heat tolerant. Genotypes HUW-213 x NW-1076 followed by HALNA x RAJ-4037 showed terminal heat tolerance as well as high yield, aided by morphological and quantitative traits.

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**Declaration-** It is certified that the article is in compliance with ethical standards and The authors declare that there is **NO** conflict of interest and their individual contribution is mentioned below.

### Author's contribution-

Conceptualization of research work and designing of experiments- Dr. Shailesh Marker.

Execution of field/lab experiments, data collection, analysis of data and interpretation- Rajesh Aggarwal

Preparation of manuscript- Ankit Phansal.

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