https://pphouse.org/ijbsm.php



IJBSM December 2022, 13(12):1440-1449 Pi

Print ISSN 0976-3988 Online ISSN 0976-4038

Natural Resource Management

DOI: HTTPS://DOI.ORG/10.23910/1.2022.3212

Genetic Component Analysis and Determination of Optimum Number of Clusters Based on Morpho-Physiological Traits in Wheat

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Research Article

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ABSTRACT

The present study was conducted at the instructional farm, Uttar Banga Krishi Vishwavidyalaya, Pundibari, Cooch Behar, West Bengal, India during the rabi season (November–March) of 2020–2021 aimed at to evaluate the performance of CIMMYT nursery (19th HTWYT) under Terai zone of West Bengal to assess genetic diversity and clustering them into optimum number of clusters using 12 morpho phenetic traits along with 02 physiological traits and also against spot blotch disease. ANOVA showed non-significant variation among the genotypes for all the 15 quantitative traits under study. The genotypes were also being screened against spot blotch disease and 29 were found highly susceptible, 14 were susceptible to highly susceptible and 06 were susceptible category whereas only the local check DBW 187 was found moderately susceptible to susceptible. The optimum number of clusters was determined by using K mean clustering algorithm which revealed optimum number of cluster I consisted of 24 wheat genotypes and Cluster II consisted 26 wheat lines. Among the two clusters, higher diversity was present in cluster I (276.67) than cluster II (249.684). Principal component analysis (PCA) for all the 15 traits revealed only five components having Eigen value >1.00. Among them PC 1 and PC 2 accounted for 36.53% and 12.05% variance respectively. Grain yield was found to be positively associated with 08 traits such as awn length, biological and grain yield, grain per spike, harvest index while negatively correlated with tiller m⁻¹, mean canopy temperature depression, AUDPC%.

KEYWORDS: Wheat, genetic divergence, K mean clustering, PCA

Citation (VANCOUVER): Poddar et al., Genetic Component Analysis and Determination of Optimum Number of Clusters Based on Morpho-Physiological Traits in Wheat. *International Journal of Bio-resource and Stress Management*, 2022; 13(12), 1440-1449. HTTPS://DOI.ORG/10.23910/1.2022.3212.

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Data Availability Statement: Legal restrictions are imposed on the public sharing of raw data. However, authors have full right to transfer or share the data in raw form upon request subject to either meeting the conditions of the original consents and the original research study. Further, access of data needs to meet whether the user complies with the ethical and legal obligations as data controllers to allow for secondary use of the data outside of the original study.

Conflict of interests: The authors have declared that no conflict of interest exists.

RECEIVED on 03rd August 2022 RECEIVED in revised form on 17th November 2022 ACCEPTED in final form on 01st December 2022 PUBLISHED on 19th December 2022

1. INTRODUCTION

The World's various wheat producing zones are classified into 12 mega-environments by CIMMYT (Rajaram et al., 1995, Braun et al., 2010). Mega-environments are geographical areas, though not necessarily contiguous, where wheat adaptation can be expected to be similar, due either to similar climatic, disease or crop-management constraints (Lantican et al., 2016). Breeding lines are distributed to these wide varieties of conditions across the world, with an emphasis on evaluating genotypes for greater adaptability and selection for specific environments (Braun et al., 1996, Samle et al., 2002).Furthermore, outstanding CIMMYTderived bread wheat lines have also been widely used in cross-breeding programmes across the world resulting in a significant increase in the yield gain in wheat in many countries (Manes et al., 2012, Sharma et al., 2012, Barkley et al., 2014). In recent studies it was found that the reappears to be more frequent overlap between mega-environments that were previously distinctly delineated, a phenomenon possibly due to climate change effects and expected to become more pronounced (Braun et al., 2010, Daryanto et al., 2016, You et al., 2014). As a result, new wheat varieties will need to be not only superior in yield potential but also having increased tolerance to drought and heat stress, better disease and pest resistance, more stable processing traits, and better nutritional qualities (Lopes et al., 2012, Mondal et al., 2020). The is one of the CIMMYT nurseries which distribute advanced breeding lines intended for heat challenged region around the globe every year.

In West Bengal wheat is mainly grown in warm subtropical rainfed or partly irrigated regions after the harvest of rice. Presence of biotic and abiotic stress factors remains a serious challenge towards sustaining high yield (Mitra et al, 2019, Maity et al, 2019). Terminal heat stress is a major concern as the crop is sown late due to late harvest of paddy. The temperature rises after February onwards which adversely affects the crop. High temperature stress adversely affects plant physiological processes; limiting plant growth and reducing grain yield (Mondal et al, 2013). Due to presence of high humidity, disease occurrence is also high. Spot blotch or foliar blight disease produced by *Bipolaris* sorokiniana (Sacc.) Shoem is one of the most serious diseases found in this region (Chowdhury et al., 2013, Kumari et al, 2018). This is a serious disease that creates tiny dark brown lesion on the leaf that quickly congeals and spread in sensitive genotypes. The severity is most prevalent in the Eastern Gangetic plains of South Asia, which encompass India, Nepal, and Bangladesh (Sharma and Duveiller 2006). In India, average yield losses owing to spot blotch have been found to be 15.5% (Dubin and van Ginkel, 1991) and 17% (Saari 1998), with grain yield losses ranging from 17.63% to 20% under favourable condition (Goel et al., 2006). Under severe infestation, however, yield loss might reach 80%

(Joshi et al., 2007). The Terai area of West Bengal, which has a high humidity level and a shorter winter season, is regarded a hotspot for spot blotch (Kumar et al., 2016).

In the present study, High Temperature Wheat Yield Trial (HTWYT) nursery developed by CIMMYT for heat stress environments was evaluated under Terai Agro climatic condition of West Bengal to address the above mentioned issues and also to select desirable genotypes for this agroclimatic condition. Genetic diversity along with optimum number of clusters was estimated among the genotypes.

2. MATERIALS AND METHODS

The research was carried out at the instructional farm, Uttar Banga Krishi Vishwavidyalaya, Pundibari, Cooch Behar, West Bengal, during the rabi season (November-March) of 2020-2021. The experimental material consisted of 50 different wheat genotypes (19th HTWYT nursery) including two checks (A and B) (Table 1). Check A

Table 1: List of wheat genotypes along with their pedigree details

Sl. No.	Genotype	Pedigree
1.	HTWYT-1	NAC/TH.AC//3*PVN/3/MIRLO/ BUC/4/2*PASTOR/5/KACHU/6/ KACHU
2.	HTWYT-2	NADI#1
3.	HTWYT 3	WBLL1*2/BRAMBLING/4/ BABAX/LR42//BABAX*2/3/ SHAMA*2/5/
4.	HTWYT 4	QUAIU#1/SUP152
5.	HTWYT 5	PBW343*2/KUKUNA/3/ PASTOR//CHIL/PRL/4/ GRACK/5/MUU/
6.	HTWYT 6	CHIBIA//PRLII/CM65531/3/ FISCAL*2/4/TAM200/ TURACO/5/
7.	HTWYT 7	WBLL1*2/BRAMBLING*2// BAVIS/3/CHYAK1/VILLA JUAREZ F2009/
8.	HTWYT 8	K A C H U / / W B L L 1 * 2 / B R A M B L I N G * 2 / 6 / BECARD#1/5/KIRITATI/4/
9.	HTWYT 9	S U P 1 5 2 / B A L # 1 * 2 / 3 / KINGBIRD#1//INQALAB 91*2/ TUKURU
10.	HTWYT 10	S U P 1 5 2 / B A L # 1 * 2 / 3 / KINGBIRD#1//INQALAB 91*2/ TUKURU

Table 1: Continue...

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S1. No	Genotype	Pedigree	S1. No	Genotype	Pedigree
11.	HTWYT 11	SUP152/BAL#1*2/3/KINGBIRD#1// INQALAB 91*2/TUKURU	31.	HTWYT 31	B O K O T A / 3 / ND643/2*WBLL1//2*BAJ#1
12.	HTWYT 12	SUP152/BAL#1*2/3/KINGBIRD#1// INQALAB 91*2/TUKURU	32.	HTWYT 32	S U P 1 5 2 / B A J # 1 / 3 / SWSR22T.B./2*BLOUK#1//
13.	HTWYT 13	SUP152/BAL#1*2/3/KINGBIRD#1// INQALAB 91*2/TUKURU	33.	HTWYT 33	WBLL1*2/KURUKU MUTUS//ND643/2*WBLL1/3/
14.	HTWYT 14	TUKUU//BAV92/RAYON/3/ FRNCLN/4/2*FRNCLN*2/ TECUE#1	34.	HTWYT 34	BORL14 SHA7//PRL/VEE#6/3/FASAN/4/
15.	HTWYT 15	ABLEU*2/BORL14			KAUZ/
16.	HTWYT 16	MILAN/KAUZ//BABAX/3/BAV92/4/ WHEAR//2*PRL/2*PASTOR/5/	35.	HTWYT 35	KACHU/SAUAL//PRL/3/KACHU/ KIRITATI
17.	HTWYT 17	KENYA SUNBIRD/2*KACHU// KFA/2*KACHU	36.	HTWYT 36	ROLF07//LALBMONO1*4/PVN/3/ BORL14
18.	HTWYT 18	BAVIS/NAVJ07//SUP152/BAJ#1	37.	HTWYT 37	NADI*2/3/EBW10 TALL#1/
19.	HTWYT 19	CHIPAK*2//SUP152/KENYA	20		WESTONIA-Rht5//NAVJ07
20	HTWYT 20	WBL 1*2/CHAPLO/6/CNDO/	38.	HIWYI 38	VIVITSI//WHEAR/3/PANDORA
20.	111 // 11 20	R143//ENTE/MEX175/3/	39.	HTWYT 39	BECARD/FRNCLN//2*BORL14
		AE.SQ/4/	40.	HTWYT 40	BECARD/FRNCLN//KACHU/
21.	HTWYT21	HEILO//MILAN/MUNIA/3/ KIRITAII/2*TRCH/4/2*KACHU/			KIRITATI/3/BOKOTA
		KIRITATI	41.	HIWYI41	A 1 1 1 L A / 3* B C N / B A V 9 2 / 3 / PASTOR/4/TACUPETO F2001*2/
22.	HTWYT 22	CHIBIA//PRLII/CM65531/3/ FISCAL*2/2/TAM200/ TURACO/5/	42.	HTWYT 42	MUNAL*2/WESTONIA/3/ WBLL1*2/BRAMBLING*2// BAVIS/4/
23.	HTWYT 23	W B L L 1*2/B R A M B L I N G*2// BAVIS*2/4/SWSR22T.B.//	43.	HTWYT 43	ROLF07*2/SHORTENED SR26
24.	HTWYT 24	BECARD/FRNCLN//BORL14	4.4		I RALOCAI ION//MUNAL#1/3/
25.	HTWYT 25	TACUPETO F2001*2/KIRITATI// BLOUK#1/3/WBLL1*2/	44.	F11 VV 1 1 44	KFA/2*KACHU
26.	HTWYT 26	QUAIU#1/BECARD/3/WBLL1*2/ BRAMBLING*2//BAVIS	45.	HTWYT 45	MUTUS*2//TAM200/TURACO*2/3/ KFA/2*KACHU
27.	HTWYT 27	B O R L 1 4 * 2 / 8 / R E H / H A R E // 2 * B C N / 3 / C R O C 1 / AE.SQUARROSA (213)/	46.	HTWYT 46	M U T U S * 2 / H A R I L # 1 * 2 / 3 / S W S R 2 2 T. B . / 2 * B L O U K # 1 / / WBLL1*2/
28.	HTWYT 28	B O R L 1 4 * 2 / 8 / R E H / HARE//2*BCN/3/CROC 1/	47.	HTWYT 47	PASTOR//HXL7573/2*BAU/3/ WBLL1/4/SOKOLL/3/PASTOR//
29.	HTWYT 29	AE.SQUARROSA (213)/ SWSR22T.B./2*BLOUK#1//WBLL*2/	48.	HTWYT 48	I S E N G R A I N / K B I R D / / MUNAL#1*2/3/KFA/2*KACHU
20		KURUKU/3/BORL14/4/	49.	HTWYT 49	CROC 1/AE.SQUARROSA (205) // BORL95/3/PRL/SARA//TSI/
50.	111 VV I I 30	MEX175/3/AE.SQ/4/2*OCI/6/ VEE/	50.	HTWYT 50	CROC 1/AE.SQUARROSA (205) // BORL95/3/PRL/SARA//TSI/

(HTWYT 1= DBW 187) was the local check and Check B (HTWYT 5) was considered as check based on yield. The farm is located at $26^{\circ}19'86$ " North latitude, $89^{\circ}23'53$ " East longitude, and is 43 metres above sea level. The genotypes were timely sown using Augmented Randomised Complete Block Design having plot size $2.5 \times 1 \text{ m}^2$ (6 rows plot⁻¹), row to row spacing 20 cm and was timely harvested. The recommended cultural practices were adopted to raise a good crop.

2.1. Study of morpho-phenetic traits

Data were recorded on five randomly selected competitive plants from each plot for 12 morpho-phenetic traits namely, germination(%) i.e. plant per meter (GER), days to 50% heading (DF), plant height in cm (PH), awn length in cm (AL), spike length in cm (SL), grain per spike (GPS), spikelet per spike (SPS), tiller per meter (TM), 1000 grain weight in gram (TGW), grain yield in t ha⁻¹ (GY), biological yield in t ha⁻¹ (BY), harvest index (HI).

2.2. Study of physiological traits

2.2.1. Canopy temperature depression

The canopy temperature was measured twice, at 68 DAS and 93 DAS, using an AR20 (Intell smart) infrared thermometer. Before recording the canopy temperature, the same infrared thermometer was used to obtain the air temperature by concentrating it on a blank sheet (white paper) positioned slightly above each plot. Canopy Temperature Depression (CTD) was calculated by subtracting the canopy temperature (T_a) from air temperature (T_a) [CTD= T_a - T_c] (Balota et al., 2008). It represents overall integrated physiological response of plants to drought and high temperature condition and thus has been used widely to assess plant response towards environmental stress. Also mean CTD (MCTD) was calculated by taking average of all the recordings.

2.2.2. Chlorophyll index

It was measured at four crop growth phases viz. 88 DAS, 95 DAS, 102 DAS, and 109 DAS using a Field Scout CM 1000 chlorophyll metre. Area under chlorophyll index progress curve (AUCIPC) was calculated as per following formula adapted from Rosyara et al., 2007.

AUCIPC
$$\sum_{i=1}^{(n-1)} 1/2 (S_{i+1} + S_i) d$$
(1)

Where, S_i =Chlorophyll index value at the end of time 'i', S_{i+1} = Chlorophyll index value at the end of time 'i+1', d = Day's interval between two observation, n = number of times of recording the value. Mean AUCIPC (MAUCIPC) was calculated by taking average of all the recordings.

2.2.3. Disease evaluation

The genotypes were screened against Spot blotch disease by creating artificial epiphytotic condition by inoculating the wheat lines with a pure culture of local isolate of *Bipolaris*

sorokiniana (Sacc) Shoem. A spore suspension of 10⁴ conidia/ml was uniformly sprayed during evening hours at tillering, flag leaf emergence and anthesis stages of the crop (Chaurasia et al., 1999). In addition, susceptible variety Sonalika was planted after every 20 lines and also in alley across the border to ensure proper inoculums in the field. The field was heavily fertilized and frequently irrigated to provide a suitable environment for the disease development.

Disease scoring was done at four crop growth stages viz. 88 DAS, 95 DAS, 102 DAS, and 109 DAS using the Double-Digit scale (Saari and Prescott 1975). The first digit (D_1) denotes disease progression from ground level to canopy height; the second digit (D_2) denotes disease severity as assessed by diseased leaf area. D_1 and D_2 are both graded on a scale of 1 to 9. For each score, the percentage of disease severity is estimated based on the following formula:

Severity (%)=
$$(D_1/9) \times (D_2/9) \times 100$$

Area under Disease Progress Curve (AUDPC) was calculated by using formula given by (Wilcoxson et al., 1975):

AUDPC=
$$\sum_{(i=1)}^{(n-1)} 1/2 (X_{i+1} + X_i)d$$
(2)

Where, X_{i+1} = Disease severity on 'i+1th day, X_i = Disease severity on 'ith day, d=Day's interval between two observations, n= number of dates on which the disease was recorded. For proper comparison among the germplasm accession, AUDPC values were standardized by maturity recorded for each genotype at each location to make it as AUDPC percent days which was determined by dividing total AUDPC by the total number of days in the evaluation period, according to Reynolds and Nehar, 1997.

2.2.4. Statistical analysis

The experimental data collected were compiled by taking the mean values of selected plants in each plot and subjected for Analysis of variance, broad-sense heritability (h²), genetic advance (GA % of mean), genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) using the R software. The h², GA% of mean, GCV and PCV were classified into high, moderate and low following the scale provided by Johnson et al., 1955. The quantitative data set were further classified into different clusters using K mean clustering algorithm in R software. The principal component analysis (PCA) and the correlation analysis were done in the PAST software.

3. RESULTS AND DISCUSSION

3.1. Mean performance of genotypes

The analysis of variance (ANOVA) (Table 2) showed non-significant variation among the genotypes for all the 15 quantitative traits under study. This indicated that the International Journal of Bio-resource and Stress Management 2022, 13(12):1440-1449

Table 2: ANOVA table of 15 quantitative traits under study									
Source	Df	GER	DF	PH	AL	SL	GPS	SPS	TM
Block (ignoring Treatments)	1	113.49	1.23	14.33	1.7	0.62	114.32	0.01	45.89
Genotype (eliminating Blocks)	49	44.1	23.21	23.17	0.96	2.06	38.49	1.85	42.52
Genotype: Check	1	0.25	25.00	166.4	0.00	10.89	299.29	3.24	9.03
Genotype: Test and Test vs. Check	48	45.01	23.17	20.18	0.98	1.88	33.05	1.82	43.22
Residuals	1	51.34	1	1.21	0.36	0.49	6.25	0.64	186.73
Table 2: Continue	Table 2: Continue								
Source	TGW	GY	BY	HI	MC	TD	MAUCIPC	AU	JDPC%
Block (ignoring Treatments)	0.79	3.3	17.74	0.0029	1.0	00	120010.65	16	5222.83
Genotype (eliminating Blocks)	12.32	0.23	2.49	0.0015	0.2	25	50809.35	3	159.67
Genotype: Check	3.53	2.15	29.27	2.5e-05	0.0)2	815679.92	-	15630
Genotype: Test and Test vs. Check	12.51	0.19	1.93	0.0016	0.2	25	34874.55	2	899.87
Residuals	3.24	0.08	7.45	0.003	0.2	23	159640.2	1	546.06

genotypes used in the present study were very much uniform in terms of the present characters studied. Mean squares due to block for all the above characters were found nonsignificant, indicating the non-adequacy of the block for statistical analysis of the characters. Mean squares due to genotypes for all the above-mentioned characters were also found non-significant which might due to their similar kind of genetic make-up. Mean squares due to checks for all the above characters along with interaction between checks and test genotypes were also found non-significant.

The coefficient of variation (CV%) was found high for traits such as TM, MCTD and BY, while moderate for GER, MAUCIPC, and HI. However, it was low for traits such as DF, PH, AL, SL, GPS, SPS, AUDPC%, TGW and GY (Table 3). The average days to 50% flowering was 66.62 days, which varied from 57.5 (HTWYT 19) to 78.5 (HTWYT 22) days. Plant height ranged from 99.25 cm (HTWYT 1) to 75.35 cm (HTWYT 17) with an average value of 86.62 cm. Among the reproductive traits AL, SL, GPS and SPS did not show much variation between genotypes. However, tillers per meter showed high variability with a range from 76.66 (HTWYT 48) to 43.17 (HTWYT 32). Highest grain yield was exhibited by HTWYT 38 (4.48 t ha⁻¹) followed by HTWYT 50 (4.23 t ha⁻¹). The lowest yield was given by HTWYT 17 (1.61 t ha⁻¹). Only six genotypes (HTWYT-28, 29, 38, 39, 49 and 50) outcrossed both Check A (3.7 t ha⁻¹) and Check B (2.23 t ha⁻¹) with respect to grain yield.

Among the physiological traits studied, MCTD values showed highest variability (58.25%) with an average value of 0.83 which varied from 2.05 (HTWYT 26) to -0.16 (HTWYT 42). The MCTD indicated gradual decline of CTD with advance in growth stages in most of the genotypes except 13 genotypes such as HTWYT 5, 8, 14, 18, 20, 21,

Table 3: Descriptive statistics of the 15 quantitative traits of 50 wheat lines

Characters	Mean	Min	Max	Std. Error	CV%		
				LIIUI			
GER	44.05	31.42	61.42	0.99	16.29		
DF	66.62	57.5	78.5	0.68	1.5		
PH	86.62	75.35	99.25	0.8	1.27		
AL	7.12	2.1	9.2	0.14	8.4		
SL	10.17	8.65	17.55	0.19	6.84		
GPS	48.5	31.05	63.65	1.37	5.14		
SPS	18.61	14.5	21.8	0.23	4.29		
ТМ	61.18	43.17	76.66	1.18	22.44		
TGW	39.71	31.7	47.38	0.5	4.52		
GY	2.85	1.61	4.48	0.11	9.63		
BY	8.76	4.86	13	0.39	30.99		
HI	0.33	0.24	0.46	0.01	16.88		
MCTD	0.83	-0.16	2.05	0.07	58.25		
MAUCIPC	2264.52	1873.53	2828.75	31.47	17.61		
AUDPC%	58.8	36.67	74.77	1.22	9.61		

24, 28, 34, 35, 46, 48 and 49 where the CTD increased at later growth stages. This indicated high physiological efficiency for these genotypes under present environmental condition. In some genotypes, such as HTWYT 1, 2, 6, 10, 15, 17, 22, 37, 39, 41, 42, 43 and 45, CTD values moved to negative one at later stages. One genotype (HTWYT 39) showed negative values inboth the growth and maturity stages . Negative CTD indicated that canopy was hot than air temperature. This might be due to either poor plant water status or less physiological efficiency. To quantify the rate in increase of Chlorophyll index value, the Area under Chlorophyll Index Progress Curve (AUCIPC) was estimated as per the aforesaid formula. Highest value of AUCIPC was shown by the Check variety (DBW 187) (2828.75) followed by HTWYT 29 (2629.68) while the lowest value was produced by genotype HTWYT 37 (1,873.53). High AUCIPC indicates higher retention of chlorophyll at maturity thus having higher chlorophyll efficiency at maturity stage. A similar result was found by Rosyara et al., 2010, where chlorophyll content was measured by SPAD reading and AUSDC (Area under SPAD Decline Curve) value was calculated after anthesis.

Evaluation of genotypes against Spot blotch [Bipolaris sorokiniana (Sacc.) Shoem] disease was done by scoring disease at four crop growth stages and then by calculating Disease severity and AUDPC (Area Under Disease Progress Curve) value as per above-mentioned formula. AUDPC% values ranged from 519.595 (HTWYT 35) to 283.095 (HTWYT 1). Genotypes were classified into resistant (R), moderately resistant (MR), moderately resistant to moderately susceptible (MR-MS), moderately susceptible (MS), moderately susceptible to susceptible (MS-S), susceptible (S), susceptible to highly susceptible (S-HS) and highly susceptible (HS) as per AUDPC values suggested by Liathikas and Ruzgas 2012. Among them, 29 were found highly susceptible (HS), 14 were susceptible to highly susceptible (S-HS) and 06 were susceptible (S) category whereas only one genotype i.e., HTWYT 1 was found moderately susceptible to susceptible (MS-S) which was a local check variety i.e., DBW 187. It indicated that all the germplasm of the present nursery was either highly susceptible or susceptible to this disease.

3.2. Genetic variability analysis

Genotypic and phenotypic coefficients of variation (GCV and PCV) were analysed and classified into three categories: low (10%), moderate (10-20%), and high (>20%) as per Singh and Chaudhary 1979. Among the 15 traits MCTD was highly influenced by the environment as the difference of PCV (63.49) with GCV (27.52) was quite high (Figure 1a). Similar results were reported for moderate GCV and high PCV for spike length (Dhananjay et al., 2012), for number of grain spike⁻¹ (Singh et al., 2013). Heritability in broad sense value, which was categorised as low (<0.3), medium (0.3-0.6) and high (> 0.6) as per Johon et al., 1955, was found high for traits such as PH, DF, AL, SL, GPS, SPS, TGW and GY (Figure 1b). This indicated that these traits are governed by additive gene action and selection may be effective for these characters. Similar result of high heritability was found in case of PH, AL and DF by earlier workers also (Nath et al., 2021). Genetic advance as a percentage of mean (GAM) was calculated according to (Falconer and Mackay 1996) and classified as low (15%), moderate (15–20%), and high (>20%), as shown in Figure



Figure 1a: Estimates of GCV and PCV of 15 traits for 50 wheat genotypes



Figure 1b: Estimates of heritability in broad sense (h²bs) of 15 traits for 50 wheat genotypes



Figure 1c: Estimates genetic advance in percent of mean of 15 traits for 50 wheat genotypes

1c. Based on the results, GAM was found high for MCTD value only.

3.3 Genetic divergence analysis

Using the K mean clustering algorithm, 50 wheat genotypes used in the present study were distributed into different number of clusters (Figure 2) based upon fifteen morphophysiological traits. The optimum number of clusters for this study was determined using Silhouette width and Gap statistics as shown (Figure 3). This determined that optimum number of cluster was 02 for the 50 wheat genotypes under study. Cluster I consisted of 24 wheat genotypes and Cluster II consisted 26 wheat lines (Table 4a). Check A and Check B are grouped in cluster I and



Optimal number of clusters silhousette width 0.25 0.20 0.15 0.10 Average 0.00 0.00 7 1 2 3 4 5 6 8 ĝ 10 Number of cluster K Optimal number of clusters <u>¥</u>0.34 0.32 0.32 0.28 0.26 2 3 4 5 7 8 9 10 11 12 13 14 15 1 6

Figure 2: Different number of clusters using K means algorithm

Figure 3: Determination of optimum clusters using Silhouette width and gap statistics

Number of cluster K

cluster II respectively. Intra cluster value was found higher than inter cluster values (Table 4b) which indicated more diversity within the cluster. Among the two clusters, higher diversity was present in cluster I (276.6687) than cluster II (249.6843). As the variability within genotypes was found non-significant, the number clusters was also found less indicating less diversity among the genotypes.

Cluster mean of the wheat genotypes (Table 4c) showed that higher GY and BY was from cluster I (3.51 t ha⁻¹ and 11.28 t ha⁻¹ respectively). Moreover, the yield contributing traits like GPS and PH were higher in cluster I (56.76 and 90.60 cm respectively). At the same time, highest chlorophyll retention value was exhibited by cluster I having highest MAUCIPC value (2382.21). In terms of disease infestation, it was also found that a lower AUDPC% value had been shown by cluster I (54.18) which indicated less disease infestation. Thus, it could be concluded that the most promising genotypes in terms of yield, physiological efficiency and disease susceptibility have been grouped in Table 4a: Distribution of 50 wheat genotypes into different clusters

Cluster No	No. of entries	HTWYT No.
I	24	1, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 49, 50
II	26	2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 48

Table 4b: Intra and Inter cluster values of 50 genotypes of wheat

Cluster no	I	II	_
I	276.6687	208.6469	
II		249.6843	

Table 4c: Cluster mean of 15 characters of wheat genotypes

Characters	Cluster I	Cluster II
GER	45.95	42.30
DF	66.52	66.71
PH	90.60	82.94
AL	7.23	7.03
SL	10.35	10.01
GPS	56.76	40.88
SPS	19.70	17.60
ТМ	55.08	66.81
TGW	39.50	39.90
GY	3.51	2.24
BY	11.28	6.43
н	0.33	0.32
MCTD	0.75	0.91
MAUCIPC	2382.21	2155.88
AUDPC%	54.18	63.06

cluster I. Cluster II had high mean value of traits such as MCTD (0.91) and TM (66.81).

3.4. Principal component analysis

Principal component analysis (PCA) is important for the reflection of the highest contributor to the total variation at each axis of differentiation (Sharma et al., 1998). PCA was done by using all the fifteen characters that were used for K mean cluster analysis. Among the fifteen principal components (PCs) only five components (PCs) had showed Eigen value >1.00, which accounted for 73.48% of cumulative proportion of variance (Table 5). Among them PC 1 and PC 2 accounted for 36.53% and 12.05% variance respectively. Scree plot was drawn to deduce the top most

Table 5: PCA summary					
PC	Eigenvalue	% variance			
1	5.48	36.528			
2	1.8	12.051			
3	1.47	9.77			
4	1.26	8.391			
5	1.01	6.73			
6	0.94	6.28			
7	0.86	5.78			
8	0.72	4.82			
9	0.44	2.95			
10	0.30	2.03			
11	0.21	1.45			
12	0.200	1.33			
13	0.19	1.24			
14	0.08	0.58			
15	0.004	0.02			

variable component, which also suggested contribution of first two components as the highest one.

The loading value of different characters has been presented in Table 6. These showed both negative and positive loadings which indicated the presence of positive and negative correlation trends between the components and the variables. Therefore, the characters which loaded high

Table 6: Five principal components along with their factor							
loadings							
Characters	PC 1	PC 2	PC 3	PC 4	PC 5		
GER	0.190	-0.370	0.101	-0.313	-0.097		
DF	0.039	0.352	0.141	-0.472	0.536		
PH	0.372	0.217	0.046	-0.087	-0.097		
AL	0.094	0.414	-0.281	-0.131	-0.287		
SL	0.114	0.177	-0.052	0.588	0.290		
GPS	0.373	-0.198	0.061	0.014	-0.018		
SPS	0.326	-0.255	0.252	0.073	0.057		
TM	-0.273	-0.001	0.113	-0.306	0.209		
TGW	-0.025	0.068	0.554	0.311	0.212		
GY	0.382	-0.076	-0.276	-0.036	0.058		
BY	0.403	-0.068	-0.032	-0.085	-0.127		
HI	-0.002	-0.080	-0.633	0.149	0.417		
MCTD	0.008	0.515	0.053	0.001	-0.335		
MAUCIPC	0.305	0.278	0.133	0.194	0.032		
AUDPC%	-0.283	-0.149	-0.024	0.212	-0.358		

positively and negatively contributed more to the diversity and they were the ones that were responsible for creating differences between clusters. In PC 1, characters such as BY, GY, GPS, SPS, MAUCIPC, and PH had high positive loadings and thus contributed positively while AUDPC% and TM had negative loadings which contributed negatively. In PC 2, AL, DF and MCTD had high positive loadings while GER and SPS had negative loading. The findings of PCA revealed that these effective contributing traits in PC 1 and PC 2 had a significant role in diversification of genotypes and selection may be possible based on this trait for future breeding programmes.

3.5. Correlation analysis

The correlation analysis (Figure 4) revealed that among the 15 characters only eight of them were positively associated with grain yield such as AL, BY, GER, GPS, HI, MAUCIPC, PH, SL and SPS. Grain yield was negatively associated with MCTD, AUDPC% and TM. Among the physiological traits MAUCIPC was found positively associated with most of the yield attributing traits such as AL, BY, GPS, GY, PH, SL and SPS. This indicated that with higher chlorophyll retention capacity of the genotype, biomass level was also increased leading to more grain yield.

However, MCTD was found no significant correlation with any of the other traits indicating less impact on overall performance of the genotypes under present environmental conditions. AUDPC% was found positively associated with only TM, whereas, negatively associated with BY, GPS, GY, MAUCIPC, PH and SPS. This indicated that higher infestation by spot blotch had negative effect on yield and yield attributes. The negative association with MAUCIPC might be due to greater loss of greenness during high disease infestation, leading to decrease in grain yield. Similar finding was found by, Malik et al., 2008, Rosyara et al., 2007, Rosyara et al., 2010.



Figure 4: Correlation matrix between 15 morpho-physiological traits of wheat

4. CONCLUSION

The genotypes were not differing significantly in terms of fifteen quantitative traits studied. However, they showed differential GCV and PCV along with variation in heritability estimates. Genetic divergence was found among the genotypes and optimum number of clusters was determined by using K mean clustering algorithm which revealed two clusters only with Cluster I having 24 wheat genotypes and Cluster II with 26 wheat genotypes.

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