## **STUDY OF GENETIC DIVERSITY OF WHEAT (***Triticum aestivum* **L.) GENOTYPES UNDER TERAI AGROCLIMATIC CONDITIONS**

### **Thesis**

Submitted to the

Uttar Banga Krishi Viswavidyalaya

In partial fulfilment of the requirement for the degree

of

### **MASTER OF SCIENCE AGRICULTURE**

in

## **GENETICS AND PLANT BREEDING**

By

## **SOURIK PODDAR**

**A-2019-036-M**



## **DEPARTMENT OF GENETICS AND PLANT BREEDING FACULTY OF AGRICULTURE UTTAR BANGA KRISHI VISWAVIDYALAYA PUNDIBARI-736165**

**2021**

## DEDICATED TO

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### *CERTIFICATE*

This is to certify that the work recorded in the thesis entitled "**Study of genetic diversity of wheat (***Triticum aestivum* **L.) genotypes under terai agroclimatic conditions**" submitted by **Mr. Sourik Poddar (A-2019-036-M)** Master of Science (Agriculture) in Genetics and Plant Breeding, Uttar Banga Krishi Viswavidyalaya is a genuine and original one carried out under my personal supervision and guidance. The results of the investigation reported in the thesis have not so far been submitted for any other Degree or Diploma. The assistance and help received from various sources during the courses of investigation is being duly acknowledged.

> (SAIKAT DAS) Chairman Advisory Committee

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Certificated that the thesis entitled "**Study of genetic diversity of wheat (***Triticum aestivum* **L.) genotypes under terai agroclimatic conditions**" submitted by **Mr. Sourik Poddar** bearing Registration Number **A-2019-036-M** towards partial fulfilment of the requirement for the award of Master Degree from the Department of Genetics and Plant Breeding, Faculty of Agriculture under Uttar Banga Krishi Viswavidyalaya has been checked against plagiarism through Urkund software on 04-08-2021 and the similarity index has been achieved as 3% which is below the maximum tolerable range as per stipulation of this Viswavidyalaya. The thesis of **Mr. Sourik Poddar** may be accepted for the award of the **Master Degree in M.Sc. (Agriculture)** of Uttar Banga Krishi Viswavidyalaya.

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## **APPROVAL OF THE EXAMINERS FOR THE AWARD OF THE DEGREE OF MASTER OF SCIENCE (AGRICULTURE) IN GENETICS AND PLANT BREEDING**

We, the undersigned, having been satisfied with the performance of Mr. Sourik Poddar, in the viva-voce examination on the Final evaluation of the thesis, conducted today, / / 2021, recommended that the thesis be accepted for the award of the degree of Master of Science (Agriculture) in Genetics and Plant Breeding.



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**Date**: Signature

Place: Pundibari, Cooch Behar. **(SOURIK PODDAR)** 

### **CONTENT**





Table 4.16. Correlation Matrix between 15 characteristics of wheat 56

### **LIST OF TABLES**



### **LIST OF FIGURES**

### **ANNEXURE**



**ABSTRACT**

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Rice-wheat cropping is the major cropping system on 13.5 mha in the Indo-Gangetic plains (IGP) (Pathak *et al.,* 2003; Panigrahy *et al.,* 2011) producing about 50% of the total food grain and feeding 40% of India population (Gupta *et al.,* 2016). Now, this rice-wheat cropping system has started showing declining trends in marginal yield due to 'heat stress' problem in last decade. Early onset of significantly higher temperature coincided with wheat grain filling stage especially in Indo-Gangetic plains leading to terminal heat stress and reduction in yield. The present investigation has been carried out with 50 advanced genotypes of wheat targeted for heat stressed areas to assess the genetic diversity of wheat genotypes for yield and yield components along with physiological attributes and also to screen the genotypes against prevailing diseases of this area.

Among the morpho-phenetic traits, only days to heading, plant height, awn length, spike length, spikelet per spike, 1000 grain weight and biological yield showed significant variation between genotypes. AUCIPC value calculated on the basis of Chlorophyll Index (CI), was highest in ENTRY 1 which indicated higher retention of chlorophyll at maturity. Canopy Temperature Depression (CTD) showed higher value for 13 genotypes at later stages which signified high physiological efficiency for these genotypes. Correlation study indicated cooler canopy temperature leads to higher grain yield and increased biomass. In terms of spot blotch resistance, no genotype was found either moderately resistant (MR) or resistant (R) category.

 $D<sup>2</sup>$  analysis showed significant diversity between genotypes and it divided 50 genotypes into 5 clusters. In terms inter cluster distance value, most divergent cluster was found as cluster I and V. Five principal components (PCs) showed Eigen value >1.00, which accounted for 69.03% of cumulative proportion of variance. Biplot curve showed, in PC 1 characters such as BY, PH, GPS, AUCIPC had high positive loadings while AUDPC and CTD had negative loadings. In PC 2, GY and HI had high positive loadings while DF had negative loading. Also, strong positive correlation was found between GY, BY, GPS while negative association between GY and DF , CTD and PH.

The correlation analysis among the 15 characters showed that only eight traits (GER, PH, AL, SL, GPS, AUCIPC, BY and HI) were positively associated with grain yield. AUCIPC was found positively correlated with PH, CTD, BY and GY. AUDPC was found to be positively correlated with HI whereas negatively correlated with DF, PH, GPS, AUCIPC and BY. This indicated that higher disease severity was negatively associated with high chlorophyll index value.

Chairman-Advisory committee Signature of student

(DR. SAIKAT DAS) (SOURIK PODDAR)

*Chapter 1 Introduction*

Bread wheat is an allohexaploid  $(2n=42)$  and it can be grown in many different environments. Although it is best adapted in cool or temperate growing conditions, it is being cultivated in many areas of the world where heat stress is a major yield limiting factor, especially during maturity stage. These areas include lowland central and peninsular India, lowland of Terai region of Nepal, Bangladesh, Thailand, southern China, Nigeria, Sudan, Bolivian lowlands and parts of Brazil and Paraguay (Dubin and Rajaram, 1996).

The above-mentioned heat stressed areas of tropical lowlands represent 9 million hectares of wheat production and can be split into lowland humid areas (e.g., Bangladesh, eastern India, Terai of Nepal and lowlands of Bolivia and Paraguay) and lowland dry areas (e.g., central and peninsular India, Nigeria and Sudan). These areas are not considered as typical wheat-growing regions, but rather warmer, non-traditional wheat zones. In many of these areas, wheat is a new crop that came with the "Green Revolution" in the 1960s and 1970s. Wheat diseases in the lowland tropical environments can be severe and require significant efforts to control. Especially in lowland humid areas where humidity is high disease problem is severe.

After China, India is the world's second-largest wheat producer, accounting for roughly a quarter of global wheat output. Wheat output in India has grown by more than six times over the last four decades, from about 12 million tonnes in 1964-65 to around 107.18 million tonnes in 2019-20. Wheat acreage has risen from 13 million hectares to about 30 million hectares over this time, while productivity has improved from 9.13  $q<sup>-ha</sup>$  to 35.0  $q<sup>-ha</sup>$ . (Source: Project Directors Report, 2019-20, IIWBR, India). This was only possible due to the import of semi-dwarf genotypes from (CIMMYT) during the 1960s which was a crucial component in India's green revolution, which saw a quantum jump in wheat yield.

Rice-wheat cropping is the major cropping system on 13.5 mha in the Indo-Gangetic plains (IGP) (Pathak *et al.,* 2003; Panigrahy *et al.,* 2011) producing about 50% of the total food grain and feeding 40% of India population (Gupta *et al.,* 2016). In the last few decades climate change is changing our globe in which agricultural crops suffers badly (Rosenzweig *et al.,* 2014). Now, this rice-wheat cropping system has started showing declining trends in marginal yield, groundwater depletion, soil deterioration and heat stress problem in last decade. Rise in temperature is one of the most important concerns as metabolic processes that eventually impact the agricultural production is governed by temperature (Teixeira *et al.,* 2013). Further increase in temperature will negatively affect agricultural production and more serious effect will be from short episodes of extremely high temperatures, or occurrence of 'heat stress'. This effect is being observed in India especially in wheat crop since last decade (Dubey *et al,* 2020). Early onset of significantly higher temperature coincided with wheat grain filling stage especially in Indo-Gangetic plains leading to terminal heat stress and reduction in yield. The possible reason behind this is the proximity to the equator and late sowing of wheat in India which exposes the crop to high temperature during grain filling stage (Joshi *et al.,* 2007).

Terminal heat stress in wheat occurs when mean temperature during grain filling stage goes above 31°C. The IPCC (2014) projected that for Indian region, temperature will increase by 0.7–2.0 °C by 2030s and 3.3–4.8 °C by 2080s. The increase may be more in north India and during the rabi season (November to March). With a rise of temperature by  $0.5-1.56$  °C by 2080–2100, adverse impact on food production will occur which will cause a loss of 10–40% in food grain production in India (Parry *et al.,* 2004; IPCC, 2007). High temperature causes adverse impact on wheat physiology restricting growth and yield. At flowering stage, heat stress causes sterility of pollen and anther leads to underdeveloped embryo which in turn reduces grain number, while heat stress during grain filling stage leads to reduced grain filling rate and in turn reduces grain weight and overall yield (Mondal *et al.,* 2013). In a study it was found that even a rise of 1°C in the mean temperature in month of March-April leads to reduction in the duration of wheat crop by seven days and yield by about 400 kg per hectare (Singh *et al.,* 2011). In a study done by International Maize and Wheat Improvement Centre (CIMMYT) the areas with lower productivity potential will have more negative impact and in future with global warming, their productivity will further decrease under heat stress condition (Joshi *et al.,* 2007). All of these findings indicate that there is a need to understand the effects of rising temperatures on wheat growth and development as well as adoption of climate smart practices (Niles *et al.,* 2015).

CIMMYT nurseries have played an important role in wheat breeding throughout the world. CIMMYT has divided the world's various wheat producing zones into many megasettings and distributed sophisticated breeding lines to a variety of conditions across the world, with an emphasis on evaluating genotypes for greater adaptability and selection for specific environments (Rajaram *et al.,* 1995; Braun *et al.,* 2010). Moreover, excellent CIMMYT- derived bread wheat lines have been widely adopted in cross-breeding programmes throughout the developing world (Braun *et al.,* 1996), substantially increasing the genetic variety of wheat cultivars in many countries (Samale *et al.,* 2002). Advanced breeding lines targeted for heat stressed areas are annually distributed to internal co-operators through High Temperature Wheat Yield Trial (HTWYT) nursery.

West Bengal is not a large wheat-growing state and it occupies around 0.2 million hectares with an annual production of 0.6 million tons. Rice –wheat is the major cropping system and the crop faces much biotic and abiotic stresses. Terminal heat stress is a major concern as the crop sown late due late harvest of paddy. Temperature rises after February onwards which adversely affects the crop. 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. This is a serious disease that creates tiny dark brown lesions on the leaf that quickly congeal 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 conditions (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).

Considering the above facts, the present investigation entitled *"*Study of Genetic Diversity of Wheat (*Triticum aestivum* L.) Genotypes Under Terai Agroclimatic Conditions*"* was undertaken with the following objectives:

- 1. To study the genetic diversity of wheat genotypes for yield and yield components along with physiological attributes.
- 2. To screen the genotypes for the resistance towards major Wheat disease like Spot Blotch.

# *Chapter 2 Review of Literature*

### **2.1. Genetic diversity**

Individuals' genetic variations with regard to different morphological characters are referred to as genetic variation. It can be used to evaluate the contribution of each character to total divergence and thus aid in the selection of superior parents for hybridization programmes.

Mahalanobis *et al.,* (1936), define statistical principles as a quantitative method for estimating genetic divergence between populations. Evolutionary forces trigger changes in the frequency of different populations, resulting in genetic divergence.

For the evaluation of genetic diversity, Rao *et al.*, (1952) proposed using  $D^2$  statistics in plant breeding. Mol *et al*., (1962) discovered that geographical distribution has little effect on genetic diversity.

Mahalanobis generalised distance calculated by statistic is used to estimate the degree of divergence between biological populations and to compute the contribution of different components to the total divergence. In the field of plant breeding, Nair and Mukharjee *et al.,* (1960) were the first to use the  $D^2$  statistic as an indicator of genetic divergence for classification.

Multivariate cluster analysis was used by Voidani *et al.,* (1993) to categorise geographical sites as well as eco geographical sub-populations based on plant characters. Due to eco-geographical variation, no clear pattern of variation among the regions was discovered.

In 121 indigenous and exotic wheat varieties, Redhu *et al.,* (1995) looked at genetic variation for nine quantitative characters. They discovered that the varieties were divided into 27 groups. The presence of significant variability in plant height, number of grains per ear, 1000 grain weight, and grain yield per plant was revealed by cluster means for different characters. They also discovered that the clustering of varieties was unrelated to their geographical origin.

Walia and Garg *et al.,* (1996) used cluster analysis to look at grain yield and its related traits in 405 pure breeding lines. They discovered 13 distinct clusters, and the clustering pattern of genotypes from the same country showed that they were distributed in several clusters,

indicating that regional and genetic variation was not parallel. Clusters IV and IX had a lot of diversity, and Cluster VI had a lot of high mean values for grain yield, biological yield, number of tillers/unit area, and harvest index.

Sharma *et al.,* (1998) investigated genetic divergence in 51 spring wheat genotypes, clustering them using three Tocher's values: 1060, 500, and 300. They discovered that genotypes were divided into seven, nine, and ten distinct clusters, respectively. At Tocher's values of 500 and 300, the linkage dendrogram and minimum spanning tree showed conformity with the clustering pattern of the  $D^2$  statistic.

Dotlacil *et al.*, (2000) stated that 120 accessions of European winter wheat land races and obsolete cultivars were clustered into eight clusters, and that clustering reduced cultivar heterogeneity within clusters in most of the evaluated characters. However, it was difficult to find a clear correlation between cultivar geographic origin and presence in specific clusters.

Bergale *et al.,* (2001) looked at genetic divergence among fifty bread wheat cultivars and found that they were clustered into 11 clusters. The appearance of genotypes from various geographical origins in a single cluster suggests that the cultivars may have an ancestral relationship. They also said that plant height was the most important factor in genetic divergence.

According to Nimbalkar *et al.,* (2002), 24 genotypes were divided into 12 clusters, with cluster III and IV having the largest and lowest intra cluster distances, respectively. Cluster VII and Cluster XII had the greatest inter-cluster size. The number of grains per spike, grain weight per spike, and number of active tillers all had a significant impact on the genetic diversity.

Using Mahalanobis  $D^2$  study, Dwivedi *et al.*, (2002) investigated the genetic divergence within 72 lines of bread wheat. The genotypes were divided into eight clusters based on  $D^2$ values, with cluster I having the most genotypes and cluster VIII having the fewest. Clusters I, III, and IV genotypes were classified as varied and had higher mean values for the most significant yield component traits.

Leilah *et al.*, (2005) conducted two field trials, over two successive winter seasons using 'Yokorarogo' cultivar to show the relationship between wheat grain yield and its components under drought condition. Principal Component Analysis (PCA) has grouped three main components accounting for 74.4% of total variation of grain yield (PC1- 33.9%, PC2- 26.8% and PC3-13.7% respectively). Cluster analysis with wheat variables was used and was found that similarity level increase as the number of cluster increases. The study proved that 100 grain weight, weight of grain/spike, harvest index and biological yield were variables most closely related to grain yield and can be considered under drought condition.

The thirty genotypes were divided into six clusters, according to Kumar *et al.,* (2009). Cluster VI has been defined for selecting parents for integrating grain yield per plant, tillers per plant, and plant height, cluster V for spike length, grains per spike, and early maturity, and cluster III for 1000 grain weight, according to cluster means.

Jaiswal *et al*., (2010) divided 300 indigenous bread wheat germplasm into twenty-three clusters, each with a different mean value for the characters under study. They also suggested that genotypes with desired values from different clusters could be used in breeding programmes to improve yield and bread wheat characters.

The 49 bread wheat genotypes were classified into 22 distinct groups by Hailegiorgis *et al.,* (2011) using cluster analysis. This suggests that the genotypes examined have a lot of variation. Cluster 9 and 13 genotypes should be considered for direct use as parents in hybridization operations to create high yielding wheat varieties based on cluster mean values. Clusters 3 and 16 genotypes can be utilised to increase protein and gluten content, early maturity, and other desired traits other than grain yield. The principal components analysis indicated that nine principal components (PCI to PC9) were responsible for almost 80% of the overall variance. It was also noticed that the tiny contribution of a few characteristics, rather than the cumulative effect of a lot of characters, was responsible for the separation of genotypes into various clusters. The results of this research may be utilised to design crosses and maximise the usage of genetic diversity and heterosis expression.

Singh *et al.*, (2014) used Tocher's and Euclidian methods of divergence to group thirteen wheat genotypes into four clusters. They also noticed that the genotypes and numbers of genotypes in each method's clusters were different.

Zaman *et al.,* (2014) studied divergence analysis in 30 drought tolerant genotypes of wheat. The result indicated presence of significant variation among the genotypes and was further classifies into six clusters. The PCA analysis revealed PC1, PC2, PC3, PC4 and PC5 where the most contributors to the variance with 30.78%, 20.11%, 17.75%, 10.93% and 7.63% as their respective values. The divergence contributing traits (days to heading, spike/ $m^2$ , and 1000 grain weight) as the most important for drought tolerance improvement through proper selection of parents. Depending upon cluster distance and cluster mean the genotypes of Cluster

II and VI could be considered as the parent materials for future drought tolerant high yielding hybridization programme.

Verma *et al.,* (2014) assessed the genetic variation of 108 bread wheat accessions from India and Australia for production and yield traits. They discovered that these genotypes were divided into eleven clusters with a distribution pattern suggesting that cluster IV (26) had the most genotypes, followed by cluster VI (22) and cluster II (12). In most cases, the inter-cluster gap was greater than the intra-cluster distance, suggesting greater genetic variability among accessions from different classes. Cluster VII and IX had the greatest inter-cluster difference (113.94), followed by VIII and X (97.72), indicating a great deal of variability within the clusters. Cluster X (13.96) had the greatest intra-cluster distance, while Cluster VII had the smallest (00.00). Cluster X genotypes had the largest mean grain yield, harvest index, and spike weight values. Perenjori, KRL 261 and KRL 283 from cluster X, and Gutha from cluster IX, can be used as possible donors for a hybridization programme to grow genotypes with high grain yields.

Salman *et al.*, (2014) separated 65 wheat accessions into six classes. Cluster 1 and cluster 4 had the most variety. This high level of diversity illustrates why prospective breeding programmes would have greater parental selection.

The 64 genotypes were grouped into nine clusters, according to Fikre *et al*., (2015). Cluster I and IX had the greatest inter cluster gap  $(D^2=5112.1)$ , followed by clusters II and IX  $(D^2=4694.4)$  and VIII and IX  $(D^2=3871.9)$ , indicating that they were genetically more divergent from one another than any other cluster. Crosses of genotypes from cluster I with cluster IX, cluster III with cluster IX, and cluster VIII with cluster IX are predicted to develop progenies with higher levels of genetic recombination and segregation.

### **2.2. Physiological study**

### **2.2.1. Canopy temperature depression (CTD)**

Under heat and drought stress, canopy temperature (CT) is a significant parameter that indicates relative resistance to terminal heat stress, and canopy temperature depression (CTD) is found to be closely associated with yield and yield attributes (Amani *et al*., 1996 and Reynolds *et al.,* 1998).

In the season, Rosyara *et al.,* (2007) investigated canopy temperature depression as a correlative indicator of spot blotch resistance and heat stress tolerance. In both heat stressed

(late sowing) and non-stressed (timely sowing) field conditions, ten genetically diverse genotypes were grown. A negative association  $(r=-0.72^*)$  was found between AUDPC per day and AUCTDPC, suggesting that AUCTDPC declines as foliar blight susceptibility increases. Based on AUCTDPC and AUDPC per day readings, genotypes may be classified as tolerant of either one or both stresses based on genetic variations for spot blotch resistance and heat stress response.

Under heat-stress conditions, Gowda *et al.,* (2011) investigated the relationship between canopy temperature depression, membrane stability, relative water quality, and grain yield in bread wheat (*Triticum aestivum*). They looked at 49 different bread wheat genotypes at three different sowing dates (15 November, 5 December, and 5 January) to see whether there was a connection between physiological parameters and grain yield in hot environment of New Delhi. The canopy temperature depression at anthesis and the canopy temperature depression at 10 days after anthesis demonstrated positive and important genotypic correlation coefficients in grain yield. Traits such as canopy temperature depression (at anthesis), canopy temperature depression (10 days after anthesis), and membrane damage both had high heritability estimates and could be used as selection criterion in stressful environments.

In South Asia, Mondal *et al*., (2013) investigated wheat earliness as a key to adaptation under terminal and continuous high temperature stress. They looked at 30 wheat genotypes in 13 different areas in South Asia, as well as two different conditions in Mexico. For DH, PH, GY, and TKW, there were significant discrepancies between Mega Environments (ME). MEI sites had higher mean GY of 5.26 t<sup>-ha</sup> and TKW of 41.8 g, compared to 3.63 t<sup>-ha</sup> and 37.4 g for MES. Early heading entries (79 days mean DH) outperformed local checks in every region, with GY of 2-11 percent higher and 40-44 g TKW. CT was linked to GY in the Mexico study, implying that cooler canopies can lead to higher GY under both normal and high temperature stress conditions.

Ray and Ahmed (2015) investigated the impact of canopy temperature on yield and grain growth of various wheat genotypes sown at various times. On November 29, sowing, canopy temperature depression was almost identical in all genotypes (mean canopy temperature depression at various stages was  $5.99^{\circ}$ C for BARI gom 26 and  $5.5^{\circ}$ C for Pavon 76). The results showed that in the 30 December sowing, BARI gom 26 had a higher mean value of canopy temperature depression (232 $^{\circ}$ C), while Pavon 76 had a lower value (0.88 $^{\circ}$ C) at various points, suggesting that BARI gom 26 had cooler canopies even under post anthesis heat stress. BARI gom 25 and BARI gom 26 continued to raise grain dry matter up to 32 days after sowing on December 30, while Pavon 76 stopped 8 days earlier, indicating that BARI gom 26 has a higher relative 1000-grain weight (96%) and grain yield (89%) than Pavon 76. Grain growth and yield were found to be greater in warmer environments while BARI gom 26 sustained a higher canopy temperature depression.

Under terminal heat stress, Jangid *et al.,* (2018) investigated 20 spring wheat genotypes for canopy temperature depression and remain green traits. The results of terminal heat were studied using delayed sowing at three separate dates: standard (S1; November 26, 2011), late (S2; December 25, 2011), and very late (S3; January 10, 2012). In comparison to Sl, the chlorophyll content in flag leaf decreased during the anthesis stage in S2 and S3 plants. The temperature of the canopy rose as plant growth progressed and sowing was delayed. Based on susceptibility index and relative yield loss, the results showed that NW 1014 was the most resistant genotype and K 91l was the most vulnerable to terminal heat stress.

### **2.2.2. Chlorophyll index (CI)**

Rice-wheat cropping is the most common cropping method in India. Due to the late harvesting of rice in this cropping method, wheat sowing is delayed. Wheat crops suffer from terminal heat stress in such conditions, and crop yields are reduced due to disruptions in plant physiological processes (Jangid and Srivastava *et al.,* 2018). Stay green characters, lower photosynthetic rate reduction, lower canopy temperature, and higher leaf conductance were found to be consistent with terminal heat resistance in wheat (Fischer *et al.,* 1998).

Handheld chlorophyll metres such as the SPAD 502, Field scout CM 1000, and others will calculate leaf greenness, which is positively associated with leaf chlorophyll material.

The abundance of various leaf pigments such as chlorophyll and carotenoid can be predicted using spectral reflectance. Changes in photosynthesis are more closely related to changes in chlorophyll content; both of these changes occur during the grain filling period, which has an effect on grain weight (Guendouza and Maamari *et al.,* 2012). The cumulative chlorophyll content of the leaves is calculated using the chlorophyll index.

Murdock *et al.,* (2004) compared both reflectance and transmittance/absorbance chlorophyll metres on wheat and found the reflectance-type metre (Field Scout CM 10003) to be very effective in assessing leaf chlorophyll content. Ambient and reflected light is used to measure the chlorophyll index (CI). Leaf greenness improved with growing nitrogen treatments applied at rates of 0, 30, 60, 90, 120, and 150 pounds of nitrogen per acre in a three-year trial on soft red winter wheat. When the R metre was used between 10 a.m. and 2 p.m., their similarity with both the T/A metre and the R metre was exceptional and nearly equal. This meant that in the case of wheat, CMI 1000 could be used to obtain an accurate estimation of leaf chlorophyll content.

Talebi *et al*., (2011) investigated the use of chlorophyll content and canopy temperature as drought tolerance measures in durum wheat (*Triticum durum* Desf.). Two criteria were used to test twenty-four durum wheat genotypes (well-watered and moisture-stressed). The genotypes with high yield often had a high chlorophyll content and a low canopy temperature in well-watered conditions. Furthermore, genotypes with a low canopy temperature can sustain high transpiration, photosynthetic rate, and yield under moisture-stressed conditions. In both environments, there was a strong positive association between chlorophyll quality and yield. The important association between canopy temperature and chlorophyll content with yield under moisture-stressed conditions could explain the potential for screening wheat genotypes for drought conditions.

Aryal and his associates (2015). Drought resistant wheat I genotypes sown at normal and late conditions were measured for chlorophyll content as an indication of spot blotch resistance. A split plot pattern of three replications was used to test 20 genotypes. Aditya, CSISA DRYT 5204, and CSISA DYRT 5205 had longer periods of staying green, higher SPAD, and lower AUDPC values, indicating that these three genotypes are suitable for late sown conditions.

### **2.3. Study on Spot blotch resistance**

Many diseases afflict the warmer parts of the world, and among them is spot blotch or foliar blight caused by *Bipolaris sorokiniana* (*Sacc*. In Sorok). Due to its widespread prevalence and growing severity, Shoem is one of the most concerning diseases in India and other South Asian countries (Joshi *et al*., 2002).

Van Ginkel *et al.,* (1998) and Rajaram *et al.,* (1998) described several spot blotch resistance sources and divided them into three groups: Latin America, China, and wheat wild relatives or alien species. Sanghai#4, Suzhoe#8, and Yangmai#6 were among the first Chinese sources of resistance used at CIMMYT. Latin American origins are mostly from Brazil, though some may have Italian ancestors. According to Mehta *et al.,* (1985), older resistant Brazilian commercial varieties are BH-1 146 and CNT-11.

Chaurasia *et al.,* (1999) tested CIMMYT and Indian gene pool spring wheat lines and found 43 lines to be resistant; CIMMYT lines were more resistant than Indian lines. Different approaches to studying seedling/adult plant resistance to spot blotch have been used, and they can be divided into two groups. The studies in the first category used a Mendelian approach involving crosses between resistant and susceptible genotypes followed by an analysis of segregation pattern, there are some reports of the presence of monogenic (Amey *et al.,*1951; Wilcoxson *et al*., 1990) and polygenic (Griffee *et al.,* 1925; Mehta *et al.,* 1985; Steffenson *et al.,* 1996) types of resistance to spot blotch disease., and the second group of research used a quantitative genetics approach with molecular markers. Another study used F1 and F2 generations derived from six crosses involving three cultivars to infer additive effects between two or three recessive genes (two each in cultivars PBW 343 and HS361, and three in RAJ 3702) Bhushan *et al.,* (2002). In two other experiments, epistatic interactions among three dominant genes were inferred (Neupane *et al.,* 2007). In the four moderately resistant cultivars Gisuz, Cugap, Chirya 1, and Sabuf, polygenic control with two or three genes providing stable and durable resistance was also registered (Velazquez *et al.,* 1994).

Joshi *et al.,* (2004) looked at the segregating generations (F3, F4, F5, and F6) of three crosses with resistant (ace. no. 8226, Mon/Ald, Suzhoe#8) and susceptible (Sonalika) parents, finding that resistance was regulated by three additive genes. When both of these experiments are considered together, a polygenic or quantitative existence of resistance can be hypothesised, which is confirmed by studies involving QTL interval mapping and GWAS.

Spot blotch is a serious concern for wheat cultivation in warmer and humid regions of the world, according to Acharya *et al.,* (2011). Disease intensity was linked to humidity, temperature, and soil nutrient levels. When the flag leaf and the leaf underneath the flag leaf become infected before the head emerges, the yield loss is the greatest.

In 2004 and 2005, Sharma *et al.,* (2006) and Duveiller *et al.,* (2006) found that spot blotch reduced grain yields by 4% to 38% and 25% to 43%, respectively. In 2004, the weight of a thousand kernel and the number of kernels per spike were decreased by 15% and 10%, respectively, and by 18% and 11% in 2005. According to the findings, the new cultivar Gautam's level of resistance to spot blotch reflects a partial success in breeding for resistance.

Rai *et al.,* (2016) investigated the physiological trait for tolerance to spot blotch (*Bipolaris sorokiniana*) in wheat genotypes (*Tricum aestivum*). Three wheat varieties were tested for physiological traits as well as spot blotch resistance at various sowing dates. In the 5th of November, AUCTPC (207.15) and AUDPC (287.38) reported significantly lower values, while HD-2967 recorded significantly higher values (278.53) The AUDPC was lower, but the yield was highest on November 5th (0.59g/m2). There was a positive correlation between canopy temperature and disease, but a negative correlation with the ability to remain green. Yield and canopy temperature were found to be negatively associated with disease, with the severity of the disease accounting for up to 59% of the difference in yield. The plant's green index is the only variable that has a positive relationship with yield. The linear relationship shows that as the stay green property increases, the canopy temperature decreases, and it accounts for about 72% of the canopy temperature rise. The results showed that delaying planting raises canopy temperature and lowers crop greenness index, resulting in an increase in disease and a decrease in yield.

Rosyara *et al.,* (2010) investigated the effects of spot blotch and heat stress on hexaploid wheat genotypes' canopy temperature depression, chlorophyll fluorescence, and chlorophyll quality. Spot blotch and heat stress are two major stresses on bread wheat caused by *Cochiliobolus sativus* (Ito and Kurib.) Drechsler ex Dastur (*Triticum aestivum* L.). In 2006 and 2007, eleven different bread wheat genotypes were tested in replicated field trials in Rampur, Nepal, under spot blotch epidemics and heat stress conditions. CTD (canopy temperature depression), CF (chlorophyll fluorescence), Chlorophyll content, percent disease leaf area, yield, and yield components were all measured. Individually, heat and spot blotch decreased CTD, CF, chlorophyll content, grain yield (GRY), and thousand kernel weights (TKW), with combined stress resulting in even greater reductions. Genotypes with lower GRY or TKW losses due to spot blotch also had lower yield losses due to heat stress or combined heat and disease stress, implying a connection between stress tolerance mechanisms. The highest values for chlorophyll content, CF, and CTD were found in genotypes with less disease.

# *Chapter 3 Materials and Methods*

Under the different headings in the chapter, the specifics of the experimental material utilised and procedures employed during the current investigation are explained.

### **3.1. Experimental site**

The research was carried out at the university instructional farm, Uttar Banga Krishi Vishwavidyalaya, Pundibari, Coochbehar, West Bengal, during the rabi season of 2020-2021. The farm is located at 26°19'86" North latitude, 89°23'53" East longitude, and is 43 metres above sea level.

### **3.2. Experimental material**

The experimental material consists of 50 different wheat (*Triticum aestivum*  L.) genotypes from the nursery sent by CIMMYT (19<sup>th</sup> High Temperature Wheat Yield Trial [19 HTWYT]. Whole list along with their pedigree is given in Table No. 3.1.

### **3.3. Meteorological features of the experimental site**

**Climatic condition**: The experimental location is located in the Sub-Himalayan terai agro-climatic zone, with average rainfall of 3000 mm from June to September. The temperature started to increase towards the end of February and peaked in April and May. Throughout the year, the relative humidity remained high, ranging from 39.86 to 97.86 %. Early rains in March-April 2021 hindered the mature crop, but late monsoon rains in October 2020 provided an advantage.

**Temperature**: At the beginning, temperature was moderate but gradually increased during maturity.

**Rainfall**: During the growing season the crop had a couple of unseasonably wet days during February.

**Humidity**: During the experiment, humidity was high to moderate, ranging from a maximum of 97.86% to a minimum of 36.86% in 2020-2021.



## **Table 3.1. List of wheat genotypes evaluated during 2020-2021**

**Experimental soil**: The experimental field's soil is from the Teesta alluvial plain group, and it's a sandy loam with low water holding capacity and moderate fertility.

Weeks	Period	Temperature( ${}^0C$ )		Rainfall	Number of		<b>Relative humidity</b>	
		Min.	Max.	(mm)		Rainy days Sunshine hours	Min.	Max.
45	Nov. 04-Nov.10	14.70	30.17	0.00	0.00	6.69	48.00	67.29
46	Nov. 11-Nov.17	13.63	31.46	0.00	0.00	7.56	42.86	60.14
47	Nov. 18-Nov.24	13.03	27.26	$0.00\,$	0.00	$\overline{5.53}$	52.29	82.71
48	Nov. $25 - Dec.01$	11.26	29.40	0.00	0.00	5.33	50.14	68.57
49	$Dec.02 - Dec.08$	11.14	28.26	0.00	0.00	5.46	49.86	67.86
50	Dec. 09 - Dec.15	13.04	24.53	0.00	0.00	2.20	67.29	87.00
51	Dec.16 - Dec. 22	8.87	24.09	0.00	0.00	3.86	56.57	85.29
52	Dec. 23 - Dec. 29	8.23	25.39	0.00	0.00	6.47	50.14	82.43
$\mathbf{1}$	Dec. 30 - Jan.05	7.79	26.56	0.00	0.00	6.56	44.00	69.57
$\mathbf 2$	Jan.06 - Jan. 12	10.44	23.40	$0.00\,$	0.00	2.43	69.43	86.43
$\mathbf{3}$	Jan.13 - Jan. 19	8.56	22.81	0.00	0.00	0.86	72.71	90.14
$\overline{\mathbf{4}}$	Jan. 20 - Jan. 26	7.69	22.00	0.00	$0.00\,$	2.39	69.57	89.00
5	Jan. $27$ - Feb. $02$	6.03	21.90	0.00	0.00	3.46	60.86	97.86
$\boldsymbol{6}$	Feb. 03 - Feb.09	6.91	27.44	0.00	0.00	6.61	39.86	74.86
$\overline{7}$	$Feb.10 - Feb.16$	9.19	27.59	0.00	0.00	5.46	41.86	84.29
$\bf 8$	Feb.17 - Feb.23	10.30	29.30	0.00	0.00	5.89	39.57	70.71
$\boldsymbol{9}$	Feb.24-Mar.02	13.59	28.29	0.00	0.00	3.09	56.71	82.29
10	Mar. 03 -Mar.09	11.99	28.97	$0.00\,$	0.00	4.23	47.29	67.14
11	<b>Mar 10 - Mar 16</b>	16.17	30.24	14.30	4.00	3.01	52.14	72.14
12	Mar.17-Mar.23	15.57	32.70	$\boldsymbol{0.00}$	0.00	5.57	41.43	67.71
13	Mar. 24-Mar.30	14.40	32.37	35.40	1.00	6.36	40.43	62.71
14	Mar.31-Apr.06	14.77	31.49	2.80	1.00	5.07	50.57	61.00
15	Apr. $07 -$ Apr. $13$	16.50	32.59	43.60	2.00	4.61	55.29	70.14
16	Apr. 14-Apr.20	17.47	30.77	73.40	4.00	4.24	61.00	69.29
$17\,$	Apr. 21-Apr.27	19.06	32.47	10.60	2.00	7.21	56.00	68.29

**Table 3.2. Meteorological data pertaining to the period of experimentation** 

[Source: Gramin Krishi Mausam Sewa (GKMS) project, UBKV, Pundibari, Cooch Behar.]

### **3.4. Experimental details**

The specifics of the experiment carried out in this study are listed in Table No. 3.3.

### **Table 3.3. Experimental details**



### **3.5. Agronomic practices.**

Before planting, the soil was bought to a fine tilth. The fertiliser was administered at a rate of 150:60:50 kg/ha of N:P:K, with half of the nitrogen applied as a basal dosage and the remainder at the time of the first irrigation. At the CRI, Boot, and Milk stages, three irrigations were administered. Irrigation in the form of a flood was used.

### **3.6. Equipment used.**

### **A) Meter scale.**

The data on plant height and spike length was collected using a standard metre scale.

### **B) Chlorophyll meter.**

The quantity of chlorophyll in the leaves was estimated using a Field scout CM 1000 chlorophyll metre (Spectrum technologies, Inc.). To determine the amount of chlorophyll in leaves, this metre detects light at wavelengths of 700 nm and 840 nm. At each wavelength, the ambient and reflected light is measured. As a result of the absorption of 700 nm light by chlorophyll a, the reflection of that wavelength from the leaf is decreased when compared to reflected 840 nm light. The wavelength of light with an 840 nm wavelength is unaffected by leaf chlorophyll concentration and may be used to estimate how much light is reflected owing to physical features of the leaf, such as the existence of a waxy or hairy surface. As soon as the trigger is pressed, the integrated lasers outline the target. The field of view is 0.434 inches (1.10 cm) in diameter at a distance of 11.2 inches (28.4 cm). The field vision expands to 7.4 inches (18.8 cm) in diameter at a distance of 72 inches (183 cm). From the observed ambient and reflected light data, a chlorophyll index value (0-999) is computed.

### **C) Infrared thermometer.**

The physiological data and canopy temperature were recorded using an AR20 (Intell smart) infrared thermometer. Using the concept of reflection, an infrared thermometer detects the temperature of the canopy. It includes laser sighting capabilities and monitors temperatures from -32°C to 380°C. By detecting the quantity of infrared radiation radiated by the object, this gadget can estimate temperature from a distance of 30cm (plant canopy).

### **3.7. Observation recorded.**

### **Morpho-phenotypic trait study.**

This experiment looked at 12 morpho-phenotypic traits, including germination per metre, days to heading, plant height, awn length, spike length, grain per spike, spikelet per spike, tiller per metre, 1000 grain weight, grain yield, biological yield, and harvest index, to see if the genotypes were stable. In each of the two replications, data was gathered from five random plants from each plot, and an average value was calculated for statistical analysis. The methods for recording observations are explained further down.

- **1. Germination per meter:** The number of seedlings germinated in any three rows within a 1 metre length are counted individually and averaged .
- **2. Days to heading:** The number of days it took 50% of the plot's plants to flower was counted and recorded.
- **3. Plant height (cm):** The height of the fully grown plant was measured from the base to the tip of the panicle.
- **4. Awn length (cm):** The awns' length was measured from the base of the awns' attachment to the spikelet to the tip.
- **5. Spike length (cm):** The spike's length was measured from its neck node to its tip.
- **6. Grain per spike:** Each spike's total number of grains was recorded, and an average was computed.
- **7. Spikelet per spike:** The total number of spikelets from each spike was counted, and the average was computed.
- **8. Tiller per meter:** The number of matured tillers produced by a metre length of line sown plants was counted and represented as tillers per metre.
- **9. 1000 grain weight (g):** Thousands of grains were counted from harvested bulk grains, and their weight was recorded in gram using any electric balance, which was accurate to two decimal points.
- **10. Grain yield (t/ha):** After harvesting, the total weight of the grains was calculated, and the average of the two replications was stated in tonnes per hectare.
- **11. Biological yield (t/ha):** The plot's mature plants were uprooted and weighted, and the results represented in tonnes per hectare.
- **12. Harvest index:** It was calculated as per following formula:

**HI= Grain yield/ Biological yield.**

### **3.8. Methods of recording physiological observations.**

**a) Canopy temperature depression:** The canopy temperature was measured twice, at 68 DAS and 93 DAS, using an infrared thermometer. The canopy temperature as well as the temperature of the air were measured in five randomly selected plants of each genotype. 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. Data was collected in the same order in all two replications, and the average values were adjusted by two decimals. Using the formula, the raw data was transformed into a usable format.

### **CTD= Air temperature – Canopy temperature.**

**b) Chlorophyll index**: At four crop growth phases, 88 DAS, 95 DAS, 102 DAS, and 109 DAS, physiological parameters such as Chlorophyll index were measured. The chlorophyll index data were collected using a Field Scout CM 1000 chlorophyll metre. The metre was pointed at target row portions using the laser guide lights, and the value acquired was instantly displayed. Readings were obtained between the hours of 10 a.m. and 2 p.m., with the sun behind the reader and the ambient light receiver unobscured. The CM 1000 metre measurements were obtained 3 to 5 feet from the canopy at  $45^{\circ}$  or 90° angles to the wheat canopy surface. Only if the ambient light intensity is more than one on a scale of 0-nine is the chlorophyll index value evaluated. The measurements are taken in a circular region of around 13-35 square inches (at 3-5 feet from the canopy), which includes a lot of plants and leaves.

Area under chlorophyll index progress curve (AUCIPC) was calculated as per following formula adapted from Rosyara *et al.,* 2007:

$$
AUCIPC = \Sigma 1/2 (S_{i+1+} S_i) d
$$

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 observations.

#### **3.9. Methods of recording disease observation (Spot Blotch).**

The disease spot blotch (*Bipolaris sorokiniana*) (Sacc) Shoem was seen at four crop growth stages: 88 DAS, 95 DAS, 102 DAS, and 109 DAS in this study. A double-digit scale (00-99) was created as a variation of Saari and Prescott's severity scale to score diseases (Saari and Prescott, 1975). The first digit (D1) denotes disease progression from ground level to canopy height; the second digit (D2) denotes disease severity as assessed by diseased leaf area. D1 and D2 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:

$$
Severity(\%) = (D1/9) x (D2/9) x 100
$$

#### **The Area Under Disease Progress Curve (AUDPC).**

To analyse the severity of the disease, Area Under Disease Progress Curve (AUDPC) was calculated by using the following formula given my Wilcoxson *et al.,* (1975). The AUDPC has no unit.

$$
AUDPC = \sum 1/2 (X_{i+1}+X_i) d
$$

Where,

 $X_{i+1}$  = Disease severity on 'i+1<sup>'th</sup> day  $X_i$  = Disease severity on 'i'<sup>th</sup> day  **d =** Day's interval between two observations.

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 Liatukas and Ruzgas, 2012. The AUDPC scale is as follows:



### **3.10. Statistical analysis.**

Statistical analysis was done by software such as R, Genstat and OPSTAT.

### **3.10.1. Analysis of variance**

For statistical analysis, the mean genotype values in each replication were utilised. To assess the importance of variance among the genotypes (treatments) for various characteristics, the data was analysed using a randomised full block design. Panse and Sukhatme (1989) detailed the procedures required in analysing the randomised full block design.

The following mathematical model was used in the analysis

$$
Y_{ij} = \mu + t_i + b_j + e_{ij}
$$

Where,  $i = 1, 2, 3, 4, \ldots, n$ , number of treatments (t)

 $\mathbf{j} = 1, 2, 3, 4, \ldots, \ldots$  number of replications (r)

 $Y_{ij}$  = Performance of i<sup>th</sup> genotype in j<sup>th</sup> replication

- $\mu$  = general mean of the population
- $t_i$  = effect of i<sup>th</sup> treatment
- $\mathbf{b}_i$  = effect of j<sup>th</sup> replication
- $\mathbf{e}_{ii}$  = random error associated with i<sup>th</sup> treatment and j<sup>th</sup> block

The partitioning of total variance, due to block, treatments and error and their expectation are given in the following table.

**Analysis of Variance (ANOVA) for RCBD**

Source of variance	df	Sum of	Mean sum of	<b>F</b> value
		squares	square	
Replication	$r-1$	SSr	<b>MSr</b>	MSr/MSe
Genotypes	$t-1$	<b>SSt</b>	<b>MSt</b>	MSt/MSe
Error	$(r-1)(t-1)$	<b>SSe</b>	<b>MSe</b>	
<b>Total</b>	$(rt-1)$			



For each character, the significance of variance among the treatment means was evaluated using the 'F' test at a 5% or 1% level of significance. It was indicated as significant if the computed 'F' value was higher than the table value, or vice versa. The critical difference (CD) was calculated to evaluate the significance of the difference between treatment means whenever the 'F' value was determined to be significant.

### $CD = SEd \times t (5\%)$  at error d.f.

Where,  $\mathbf{t} = \text{table value of 't' at error d.f.}$ 

**SEd** = standard error of difference between two treatment means
$$
\mathbf{SEd} = \sqrt{2MSe/r}
$$

Where, **MSe** = Mean sum square of error

**r** = Number of replications.

If the difference between the two-treatment means is higher than the CD value (at 5% or 1%), it is said to be significantly different or visa-versa

#### **3.10.2. Heritability and Genetic advance**

## **Heritability**

Heritability in broad sense *h²* (b) was computed as a ratio of genotypic variance to phenotypic variance (Allard, 1960)

$$
h^2\left(\mathbf{b}\right) = \frac{\sigma^2 g}{\sigma^2 \mathbf{p}} \times 100
$$

Were,  $h^2$  (b) = Heritability in broad sense

**σ²g** = Genotypic variance

**σ²p** = Phenotypic variance

### **Genetic advance**

The expected genetic advance under selection for the different characters was estimated as suggested by Allard (1960).

$$
G.A. = h^2(b) \times \sigma_p \times k
$$

Where,

 **G.A.** = Expected genetic advance

 $h^2$ <sup>b</sup> = Heritability in broad sense

 $\sigma_{\rm p}$  = Phenotypic standard deviation ( $\sqrt{\sigma_{\rm p}^2}$ )

 $K =$  Intensity of selection, the value of which is 2.06 when 5 percent of the individual are selected from the population as given by Lush (1949).

Genetic advance as percent of mean for each character was calculated as suggested by Johnson *et al.* (1955).

$$
Genetic advance as percent of mean = \frac{GA}{Mean} \times 100
$$

# **3.10.3. Coefficient of variance**

Following Burton and Devane (1953), the genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), and environmental coefficient of variation (ECV) were calculated.

GCV = 
$$
\frac{\text{Genotypic standard deviation } \sqrt{\sigma_g^2}}{\text{Mean}}
$$
 x 100  
Phenotypic standard deviation  $\sqrt{\sigma_p^2}$   
PCV = 
$$
\frac{\text{Phenotypic standard deviation } \sqrt{\sigma_p^2}}{\text{Mean}}
$$
 x 100

The estimates of genotypic and phenotypic standard deviations were obtained from the respective analysis of variance table for different characters. holypic a

For convenience following classifications were used for describing various parameters of variability in the text: Environme



The range of heritability and genetic advance as percentage of mean was classified as suggested by Johnson *et al*. (1955).

## **3.10.4. Estimation of Correlation coefficient**

Correlation was calculated as the relationship between different character pairings (between two variables). As proposed by Searle (1961), the genotypic association was evaluated using analysis of variance and covariance.

#### **Genotypic correlation between character x and y**

$$
r_{xy}(g) = \frac{Cov_{xy}(g)}{\sqrt{Var_x}(g) \times Var_y(g)}
$$

Were,  $\bf{Cov}_{xy}(\bf{g})$  = Genotypic covariance between two-character x and y

 $\textbf{Var}_x(g)$  = Genotypic variance for characters x

**Var**<sub>y</sub> (g) = Genotypic variance for characters y

The significance of correlation coefficient (r) was tested by comparing the observed value of correlation coefficient with the tabulated value for (n-2) degrees of freedom. If the observed value is more than the table value, the correlation coefficient is said to be significant.

$$
tc = \frac{r\sqrt{n-2}}{\sqrt{1-r^2}}
$$

Where,  $\mathbf{r} = \text{correlation coefficient}$ 

**n** = number of genotypes

**tc** = t calculated

# **3.10.5. Genetic Diversity.**

The generalised distance between two population is defined by Mahalanobis (1936) as

$$
D^2 = \sum \lambda i. j. \text{di.dj}
$$



In the current investigation, estimating  $D^2$  values from the above formula is extremely difficult since it requires inverting a thirteenth order determinant and then evaluating B(BH) /2

terms whose total equals  $D^2$ . Working using a collection of uncorrelated characters derived from the original measurements was found to be more convenient.  $D^2$  is simplified to the evaluation of a simple sum of uncorrelated character derived from original measurements when such converted variables are used.  $D^2$  is simplified to a simple sum of squares evaluation when such modified variables are used. The pivotal condensation approach was used to transform the data (Singh and Chaudhary, 1977). The coefficients for the transformation were obtained by dividing the first row of the reduced matrix by the square root of the corresponding pivotal condensation elements.

#### **3.10.6. Determination of group constellations**

Because a cluster is a poorly defined concept, no guidelines can be established for locating them. The only requirement seems to be that any two groups belonging to the same cluster should have a lower  $D^2$  score on average than two groups belonging to separate clusters. The genotypes were classified using Tocher's technique (Rao, 1952).

#### **3.10.6.1. Average intra cluster distance**

The intra cluster distance was calculated at  $\sum D^{2/n}$ ; where,  $\sum D^{2}$  is the sum of distances between all possible combinations (n) of the population included in a cluster.

## **3.10.6.2. Average inter-cluster distance**

The average inter cluster distance was calculated as  $\sum_{i}D_i/(n_in_i)$  where  $n_i$  Is the number of populations in cluster-I and  $n_i$  is the number of populations in cluster j.

## **3.10.6.3. Cluster mean**

Cluster means were calculated for individual characters on the basis of mean performance of the genotypes included in that cluster.

# **3.10.6.4. Cluster diagram**

With the help of  $D^2$  values between the clusters, a diagram showing the relationship between different populations was drawn.

## **3.10.6.5. Relative contribution of characters towards genetic diversity**

With the help of  $D^2$  statistics, the relative contribution of each component traits to the total divergence was worked out.

#### **.10.7. Principal component analysis:**

The variables under investigation are frequently highly linked, and as a result, they effectively communicate the same thing. The original collection of variables can be transformed into a new set of uncorrelated variables known as principal components. These new variables are linear combinations of original variables that are derived in decreasing order of significance, with the first principal component accounting for as much variance in the original data as feasible. PCA is also a linear dimensional reduction approach that finds orthogonal directions of maximum variance in the original data and projects it into a lowerdimensional space made up of a subset of the highest variance component.

Let  $X_1, X_2, X_3, \ldots, X_p$  are the variables under study, then first principal component may be define as

 $Z_1 = a_{11}X_1 + a_{12}X_2 + \ldots + a_{1p}X_p$ 

Such that variance of Z1 is as large as possible subject to the condition that

 $a^{2}_{11} + a^{2}_{12} + \dots + a^{2}_{1p} = 1$ 

This constraint is introduced because if this is not done, then var  $(Z1)$  can be increased simply by multiplying any a1js by a constant factor. The second principal component define as

$$
Z2 = a_{21}X_1 + a_{22}X_2 + \ldots + a_{2p}X_p
$$

Such that var  $Z2$ ) is as large as possible to var  $(Z_1)$  subject to the constraint that

A 2 <sup>21</sup> +a<sup>2</sup> <sup>22</sup> +………a<sup>2</sup> 2p =1 and cov (Z1,Z2) = 0 and so on.

*Chapter 4 Result and Discussion*

## **4.1. Study on Morpho-phenetic traits**

#### **4.1.a. Analysis of variance (ANOVA).**

The ANOVA for the 12 characters indicated significant differences between the genotypes for 50% Heading, plant height, awn length, spike length, spikelet per spike, 1000 grain weight and biological yield. This revealed that there was substantial variability present among the wheat genotypes for those traits (Table 4.1). Replications were also found to be differed significantly for traits such as plant height, spikelet per spike, tiller per meter, 1000 grain weight, biological yield and harvest index. This indicates the correct choice of design for statistical analysis.

#### **4.1.b. Mean performance along with different genetic components**

The mean values for the characters were produced in the Table 4.2 whereas the other genotypic parameters were given in Table 4.3.

## **Germination per meter**

The best performance for germination per meter was exhibited by ENTRY 3 (65) and ENTRY 2 (62.5). The lowest performance was exhibited by ENTRY 32 (33.5), ENTRY 35 (33.5), ENTRY 33 (34.65), ENTRY 26 (35), ENTRY 18 (35.65), ENTRY 31 (36), ENTRY 41 (36), ENTRY 29 (36.84), ENTRY 34 (37.17), ENTRY 21 (38.67) and ENTRY 6 (39.84). CV is moderate for this trait (19.75%) while heritability (0.089) and genetic advance (3.78%) is poor for this trait.

#### **Days to 50% heading**

The highest days to 50% heading was exhibited by ENTRY 22 (78), ENTRY 21 (76.5), ENTRY 36 (76.5), ENTRY 39 (74), ENTRY 29 (73), ENTRY 25 (72), ENTRY 26 (72), ENTRY 34 (72), ENTRY 1 (71), ENTRY 7 (71), ENTRY 14 (71), ENTRY 15 (71), ENTRY 28 (71) and ENTRY 45 (70). The shortest time required was by ENTRY 19 (58), ENTRY 31 (58) and ENTRY 11 (58.5). CV is found to be low (4.71%) while heritability (0.645) is quite high and genetic advance (10.5%) is moderate for this trait.

<b>Source</b>	df	<b>Mean sum of Square</b>											
	<b>GER</b>	DF	PH	AL	SL	<b>GPS</b>	<b>SPS</b>	TM	<b>TGW</b>	<b>GY</b>	<b>BY</b>	$\mathbf{H}$	
<b>Replication</b>	$\mathbf{1}$	13.915	5.760	996.288**	0.014	1.000	37.450	$9.120*$	643.470**	$151.610**$	0.000	$61.202**$	$0.121***$
Genotype	49	90.434	45.644**	38.959**	$1.957***$	$3.752*$	60.970	$3.386^{**}$	79.230	$24.124***$	0.437	$3.628***$	0.007
<b>Error</b>	49	75.696	9.862	18.589	0.227	2.334	42.660	1.430	65.650	8.248	0.400	1.917	0.006

**Table 4.1. : Analysis of variance of morpho phenetic traits**

**\***significant at 5% probability level, **\*\***significant at 1% probability level.

GER= Germination per meter, DF= 50% Heading, PH= Plant Height (cm), AL= Awn Length (cm), SL= Spike Length (cm), GPS= Grain Per Spike, SPS= Spikelet Per Spike, TM= Tiller per Meter, **TGW**= 1000 Grain Weight (g), **GY**= Grain Yield (t-ha), **BY**= Biological Yield (t-ha), **HI**= Harvest Index

<b>Genotypes</b>	<b>GER</b>	DF	PH	AL	SL	<b>GPS</b>	<b>SPS</b>	TM	<b>TGW</b>	GY	BY	H <sub>I</sub>
$\mathbf{1}$	42.33	71	98.3	7.6	13.3	61.5	20.7	55.17	43.01	3.448	12.770	0.267
$\boldsymbol{2}$	62.5	66	90	7.1	10.1	56.1	21.6	69.335	40.65	4.036	12.030	0.337
$\mathbf{3}$	65	66	91.7	6.8	9.4	60.6	19.8	66.335	36.7	3.654	10.450	0.353
$\overline{\mathbf{4}}$	45.5	66	93.8	8.3	10	49.1	19.5	58.665	39.58	3.644	12.070	0.309
5	41.835	66	85.4	7.6	10	44.2	18.9	52.165	41.13	3.380	7.360	0.492
6	39.835	64	84.4	7.1	9.4	52.6	17.3	57.17	37.07	3.018	9.420	0.336
$\overline{\mathbf{7}}$	45.5	71	92.7	7.4	11.2	48.5	19.1	59.5	37.51	3.654	9.380	0.396
${\bf 8}$	44.835	63	89.1	7.8	10.2	54.2	18.6	57.165	44.49	2.996	8.090	0.371
$\boldsymbol{9}$	41.335	67	89.2	7.6	10.3	54.7	20.1	60.835	44.38	3.820	9.400	0.409
10	52.5	61.5	85.6	6.4	9.5	49.9	21	64.33	39.29	3.278	8.370	0.394
11	56.17	58.5	87.5	6.3	8.9	45.9	18.3	57.165	42.17	3.594	8.920	0.403
12	42.17	61.5	82.1	6.6	10.3	43.4	17.8	67.5	46.81	3.080	8.300	0.372
13	42.17	61	83.2	7.4	9.3	49.4	18.5	65	35.75	3.738	8.010	0.476
14	44.5	71	89.6	8.5	8.9	43.3	15.6	60.67	37.36	2.982	8.450	0.352
15	36.665	71	84.6	7.3	9.3	50.2	18.3	57.835	36.47	3.286	8.690	0.380
16	41.665	65.5	85.1	$8\,$	10.1	46.5	17.4	61.165	45.84	3.246	7.720	0.425
17	45.83	62	79.2	6.6	9.3	43	17.3	72.335	43.89	2.938	8.280	0.354
18	35.665	62	83.6	7.8	9.7	40	15.4	62.5	38.75	3.768	8.790	0.437
19	44.665	58	85.3	7.3	17.7	54.9	18.7	58	39.72	4.302	8.760	0.503
20	42.83	66.5	85.6	6.9	10.5	46.1	19.6	72.165	38.33	3.848	9.030	0.429
21	38.67	76.5	86.4	6.7	9.8	44.7	17.7	61	39.86	2.546	8.460	0.299
22	46.5	78	85.2	$\tau$	9.4	44.8	17.4	62	39.69	3.036	7.800	0.398
23	47	66	80.8	7.8	11.5	50.9	19.2	57.665	35.94	3.400	9.430	0.361
24	51.165	68	91.4	7.3	10.1	54.4	19.1	66	34.73	3.808	9.920	0.384
25	42.665	72	82.3	7.3	9.9	51.8	17.2	72.165	37.3	3.434	9.100	0.367
26	35	72	91.1	7.3	10.4	45.3	18.8	62.335	36.32	3.224	9.480	0.357
27	44	67	95.4	6.9	9.8	48.8	19.4	54.665	37.48	3.434	9.640	0.366

**Table 4.2. : Mean values of morpho phenetic traits**



**GER**= Germination per meter, **DF**= 50% Heading, **PH=** Plant Height (cm), **AL**= Awn Length (cm), **SL**= Spike Length (cm), **GPS**= Grain Per Spike, **SPS**= Spikelet Per Spike, **TM**= Tiller per Meter, **TGW**= 1000 Grain Weight (g), **GY**= Grain Yield (t-ha), **BY**=Biological Yield (t-ha), **HI**= Harvest Index.

### **Plant height (cm)**

The best performance was exhibited by ENTRY 1 (98.3), ENTRY 27 (95.4), ENTRY 4 (93.8), ENTRY 28 (93.5) and ENTRY 34 (93.3). The shortest was ENTRY 17 (79.2), ENTRY 48 (80), ENTRY 37 (80.7) and ENTRY 23 (80.8). CV is found to be low (4.98%) while heritability (0.354) is medium and genetic advance (4.52%) is poor for this trait.

#### **Awn length (Cm)**

The best performance was shown by ENTRY 50 (8.9), ENTRY 14 (8.5), ENTRY 34  $(8.4)$ , and ENTRY 4  $(8.3)$ . The shortest was ENTRY 37  $(1.8)$ . CV is found to be low  $(6.68\%)$ while heritability (0.792) is quite high and genetic advance (23.95%) is high for this trait.

#### **Spike length (Cm)**

The best performance was given by ENTRY 19 (17.7). The shortest was ENTRY 48 (8.5), ENTRY 45 (8.8), ENTRY 11 (8.9) and ENTRY 14 (8.9). CV is found to be high (15.02%) while heritability (0.233) is low and genetic advance (8.23%) is low for this trait.

#### **Grain per spike**

The genotypes showing highest grain per spike are ENTRY 1 (61.5) and ENTRY 3 (60.6). The lowest grain per spike is shown by ENTRY 38 (36), ENTRY 41 (36.8) and ENTRY 40 (39.2). CV is found to be high (13.47%) while heritability (0.177) is low and genetic advance (5.4%) is low for this trait.

#### **Spikelet per spike**

The highest spikelet per spike was shown by ENTRY 2 (21.6), ENTRY 10 (21), ENTRY 37 (20.9), ENTRY 1 (20.7), ENTRY 36 (20.5) and ENTRY 9 (20.1). The lowest spikelet per spike is shown by ENTRY 18 (15.4), ENTRY 14 (15.6) and ENTRY 48 (16.1). CV is found to be low (6.43%) while heritability (0.406) is medium and genetic advance (6.98%) is low for the trait.

#### **Tiller per meter**

The maximum tiller per meter was shown by ENTRY 48 (76.34), ENTRY 17 (72.34), ENTRY 20 (72.17) and ENTRY 25 (72.17). The minimum tiller per meter was shown by ENTRY 32 (47.5) and ENTRY 31 (48.17). CV is found to be high (13.24%) while heritability (0.094) and genetic advance (2.69%) is low for the trait.

# **1000 grain weight (g)**

The highest 1000 grain weight was given by ENTRY 12 (46.81), ENTRY 16 (45.84), ENTRY 44 (44.99), ENTRY 45 (44.57), ENTRY 8 (44.49), ENTRY 9 (44.38) and ENTRY 29 (44.19). The lowest 1000 grain weight was given by ENTRY 39 (32.27). CV is found to be low (7.23%) while heritability (0.49) is medium and genetic advance (10.24%) is moderate for the trait.

# **Grain yield (t/ha)**

The highest grain yield was given by ENTRY 19 (4.302) and ENTRY 2 (4.036). The lowest grain yield was provided by ENTRY 35 (2.3), ENTRY 46 (2.438) and ENTRY 45 (2.482). CV is found to be quite high (20.38%) while heritability (0.044) is low and genetic advance (1.82%) is low for this trait.

# **Biological yield (t/ha)**

The highest biological yield was provided by ENTRY 1 (12.77), ENTRY 4 (12.07) and ENTRY 2 (12.03). The lowest biological yield was provided by ENTRY 32 (6.33), ENTRY 45 (6.56), ENTRY 41 (6.72), ENTRY 33 (6.77) and ENTRY 40 (6.88). CV is found to be high (15.81%) while heritability (0.309) is medium and genetic advance (12.08%) is moderate for the trait.

# **Harvest index**

The highest harvest index was provided by ENTRY 32 (0.553) and ENTRY 19 (0.503). The lowest harvest index was provided by ENTRY 1 (0.267), ENTRY 35 (0.270) and ENTRY 21 (0.299). CV is quite high (21.78%) while heritability (0.077) is low and genetic advance (3.36%) is low for this trait.

S. No.	<b>Character</b>	<b>Mean</b>	Range	C.V	P.C.V	G.C.V	$H^2_{\ \mathrm{bs}}$	$GA\%$
$\mathbf{1}$	<b>GER</b>	44.05	33.50-65.00	19.75	20.69	6.16	0.089	3.78
$\overline{2}$	<b>DF</b>	66.62	58.00-78.00	4.71	7.91	6.35	0.645	10.5
$\overline{\mathbf{3}}$	PH	86.62	79.20-98.30	4.98	6.19	3.68	0.354	4.52
$\overline{\mathbf{4}}$	AL	7.12	1.80-8.90	6.68	14.68	13.06	0.792	23.95
5	SL	10.17	8.50-17.70	15.02	17.15	8.28	0.233	8.23
6	<b>GPS</b>	48.5	36.00-61.50	13.47	14.84	6.24	0.177	5.4
$\overline{7}$	<b>SPS</b>	18.61	15.40-21.60	6.43	8.34	5.31	0.406	6.98
8	<b>TM</b>	61.18	47.50-76.34	13.24	13.91	4.26	0.094	2.69
$\boldsymbol{9}$	<b>TGW</b>	39.71	32.27-46.81	7.23	10.13	7.1	0.49	10.24
<b>10</b>	<b>GY</b>	3.24	2.30-4.30	20.38	19.97	4.2	0.044	1.82
11	BY	8.76	6.33-12.77	15.81	19.01	10.56	0.309	12.08
12	$\mathbf{H}$	0.38	$0.27 - 0.55$	21.78	21.22	5.88	0.077	3.36

**Table 4.3. : Genetic parameters for different morpho-phenetic characters of 50 genotypes of wheat**

**GER**= Germination per meter, **DF**= 50% Heading, **PH**= Plant Height (cm), **AL**= Awn Length (cm), **SL**= Spike Length (cm), **GPS**= Grain Per Spike, **SPS**= Spikelet Per Spike, **TM**= Tiller per Meter, **TGW**= 1000 Grain Weight (g), **GY**= Grain Yield (t-ha), **BY**= Biological yield (t-ha), **HI**= Harvest Index. **CV**= Coefficient of Variance, **PCV**= Phenotypic correlation Coefficient, GCV= Genotypic Correlation coefficient,  $H^2$ <sub>bs</sub>= Heritability (broad sense), GA%= Genetic advance as % of mean.

# **4.2. Study on physiological characters**

# **4.2.a. Chlorophyll Index (CI)**

The Chlorophyll index studies at four different crop growth stages (88 DAS, 95 DAS, 102 DAS and 109 DAS) showed significant difference among the genotypes as well as growth stages. However, genotype x growth stages interaction was found non-significant (Table 4.4a). The mean value of CI indicated gradual decline of chlorophyll index with advances in growth stages (Table 4.4c). This might be due to gradual decay in chlorophyll pigmentation with the maturity of crop. However, the rate of decrease in CI value was found highly variable among the 50 genotypes under study.

**Table 4.4a: Two-way Analysis of variance of Chlorophyll Index values** 

<b>Source of variation</b>	df	<b>Mean sum of square</b>	F pr.
Genotype	49	1629.00*	0.013
Growth stage	3	112427.00***	< 0.001
Genotype x Growth stage	147	962.00	0.636
Error	199	1016.00	

**Table 4.4b: Statistical parameters of Chlorophyll Index values**



In the first growth stage the maximum CI value is shown by ENTRY 10 (234.800) whereas the minimum value is shown by ENTRY 5 (134.300). In the second growth stage the maximum value is shown by ENTRY 1 (218.800) whereas minimum by ENTRY 37 (125.5). In the third growth stage the maximum value shown by ENTRY 28 (202.700) whereas minimum vale by ENTRY 48 (121). In the fourth growth stage the maximum value is shown by ENTRY 27 (177.800) and minimum value by ENTRY 48 (79.6). Fig 1 is the graphical representation of the Chlorophyll Index along with the growth stages.

<b>GENOTYPE</b>	<b>88 DAS</b>	<b>95 DAS</b>	<b>102 DAS</b>	<b>109 DAS</b>
1	181.900	218.800	192.300	155.400
$\overline{2}$	171.100	182.500	158.600	104.900
3	166.800	199.100	150.500	86.200
$\overline{\mathbf{4}}$	178.600	195.000	135.100	99.700
5	134.300	144.900	132.300	94.400
6	181.200	169.300	131.200	82.300
$\overline{7}$	161.900	162.700	142.200	102.800
8	207.800	182.900	169.000	113.800
9	193.900	171.300	158.900	114.000
10	234.800	169.100	127.600	91.200
11	199.500	178.500	160.200	104.400
12	178.700	171.700	159.700	94.900
13	176.000	158.700	126.400	86.100
14	192.800	170.200	154.200	126.800
15	187.700	150.200	151.800	99.400
16	166.100	197.900	153.500	92.500
17	216.900	152.700	165.000	104.200
18	164.300	145.500	146.500	105.200
19	206.700	199.400	156.900	92.800
20	182.700	163.100	148.700	93.100
21	214.400	156.500	162.000	116.100
22	146.900	146.400	137.600	102.100
23	159.100	174.600	147.500	98.400
24	171.400	176.900	121.600	85.100
25	165.200	135.700	137.900	108.500
26	171.500	168.500	170.500	108.000
27	165.400	209.800	153.400	177.800
28	202.400	169.600	202.700	133.300
29	188.600	205.700	168.000	120.400
30	168.800	149.300	143.300	108.000
31	151.900	148.200	130.100	88.600
32	205.200	168.200	148.400	98.300
33	159.400	145.500	161.300	95.000
34	183.400	163.400	147.200	104.000
35	188.500	183.000	144.300	87.700
36	199.200	146.800	194.600	122.100
37	151.500	125.500	132.000	95.800
38	161.700	178.900	130.400	88.800
39	169.100	165.800	178.700	121.800
40	176.300	163.100	143.600	86.700
41	154.600	152.000	139.100	117.500
42	162.900	149.800	128.100	108.900
43	220.400	170.500	148.800	81.300
44	165.000	149.000	165.200	97.700
45	174.500	146.900	136.800	96.400
46	160.900	150.300	163.400	95.900
47	145.300	155.300	126.900	92.800
48	159.300	146.200	121.000	79.600
49	162.600	150.800	122.200	94.800
50	170.000	178.200	127.000	87.600
Mean	177.182	166.278	149.084	102.862

**Table 4.4c. : Two-way mean table for Chlorophyll Index under different growth stages**





To quantify the rate in decrease of CI value Area Under Chlorophyll Index Progress Curve (AUCIPC) was estimated as per formula given by Rosyara *et al.,* (2007). Highest value of AUCIPC was shown by the genotype ENTRY 1 (2828.75) while the lowest value was produced by genotype ENTRY 37 (1,867.57) [Table 4.5]. High AUCIPC indicates higher retention of chlorophyll at maturity. Thus, genotypes such as ENTRY 1, 8, 19, 27, 28 and 29 were having high chlorophyll efficiency at maturity stage. 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 found significant among genotypes after anthesis.

Genotype	<b>AUCIPC 1</b>	<b>AUCIPC 2</b>	<b>AUCIPC 3</b>	<b>Mean AUCIPC</b>
1	3,806.65	3,288.80	1,390.80	2,828.75
$\boldsymbol{2}$	3,359.20	2,728.80	1,054.00	2,380.67
3	3,476.05	2,796.80	946.8	2,406.55
$\overline{\mathbf{4}}$	3,549.20	2,640.80	939.2	2,376.40
5	2,652.40	2,217.60	906.8	1,925.60
6	3,329.75	2,404.00	854	2,195.92
7	3,083.70	2,439.20	980	2,167.64
${\bf 8}$	3,711.65	2,815.20	1,131.20	2,552.68
$\boldsymbol{9}$	3,469.40	2,641.60	1,091.60	2,400.87
10	3,837.05	2,373.60	875.2	2,361.95
11	3,591.00	2,709.60	1,058.40	2,453.00
12	3,328.80	2,651.20	1,018.40	2,332.80
13	3,179.65	2,280.80	850	2,103.49
14	3,448.50	2,595.20	1,124.00	2,389.24
15	3,210.05	2,416.00	1,004.80	2,210.28
16	3,458.00	2,811.20	984	2,417.74
17	3,511.20	2,541.60	1,076.80	2,376.54
18	2,943.10	2,336.00	1,006.80	2,095.30
19	3,857.95	2,850.40	998.8	2,569.05
20	3,285.10	2,494.40	967.2	2,248.90
21	3,523.55	2,548.00	1,112.40	2,394.65
22	2,786.35	2,272.00	958.8	2,005.72
23	3,170.15	2,576.80	983.6	2,243.52
24	3,308.85	2,388.00	826.8	2,174.55
25	2,858.55	2,188.80	985.6	2,010.98
26	3,230.00	2,712.00	1,114.00	2,352.00
27	3,564.40	2,905.60	1,324.80	2,598.27
28	3,534.00	2,978.40	1,344.00	2,618.80
29	3,745.85	2,989.60	1,153.60	2,629.68
30	3,021.95	2,340.80	1,005.20	2,122.65
31	2,850.95	2,226.40	874.8	1,984.05
32	3,547.30	2,532.80	986.8	2,355.64
33	2,896.55	2,454.40	1,025.20	2,125.38
34	3,294.60	2,484.80	1,004.80	2,261.40
35	3,529.25	2,618.40	928	2,358.55
36	3,287.00	2,731.20	1,266.80	2,428.34
37	2,631.50	2,060.00	911.2	1,867.57
38	3,235.70	2,474.40	876.8	2,195.64
39	3,181.55	2,756.00	1,202.00	2,379.85
40	3,224.30	2,453.60	921.2	2,199.70
41	2,912.70	2,328.80	1,026.40	2,089.30
42	2,970.65	2,223.20	948	2,047.28
43	3,713.55	2,554.40	920.4	2,396.12
44	2,983.00	2,513.60	1,051.60	2,182.73
45	3,053.30	2,269.60	932.8	2,085.24
46	2,956.40	2,509.60	1,037.20	2,167.73
47	2,855.70	2,257.60	878.8	1,997.37
48	2,902.25	2,137.60	802.4	1,947.42
49	2,977.30	2,184.00	868	2,009.77
50	3,307.90	2,441.60	858.4	2,202.63
SE(m)	382.2	338.3	101.5	245.4
L.S.D. (0.05)	1086.2	961.4	288.3	697.3

**Table 4.5. : AUCIPC values pertaining to different genotypes under study**

# **4.2.b. Canopy Temperature Depression (CTD)**

Canopy Temperature Depression studied at two different growth stages (68 DAS and 93 DAS) showed significant difference among the growth stages. However, genotypes and genotype x growth stage interaction were found non-significant (Table 4.6a). The mean values of CTD (Table 4.6c) indicated gradual decline of CTD with advance in growth stages in most of the genotypes except 13 genotypes such as ENTRY 5, 8, 14, 18, 20, 21, 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 ENTRY 1, 2, 6, 10, 15, 17, 22, 37, 39, 41, 42, 43 and 45 the CTD values even moved to negative one at later stages. One genotype (ENTRY 39) had negative values in both the growth stages (-0.050 in 68 DAS and -0.270 in 93 DAS). This might be due to high canopy temperature due to either poor plant water status or less physiological efficiency. Fig 2 is the graphical representation of CTD along with its growth stages. Similar results were found by earlier workers such as Guendouz *et al.,* 2012 using ten durum wheat varieties to show CTD as an indicator to drought tolerance in semi-arid conditions.

**Table 4.6a. : Two-way Analysis of variance for Canopy Temperature Depression under different growth stages**

<b>Source of variation</b>	df	Mean sum of square	F pr.
Genotype	49	1.064	0.598
Growth stage		20.840***	< 0.001
Genotype x Growth stage	49	0.929	0.784
Error	199	1.140	







# **Table 4.6c. : Two-way mean table for Canopy Temperature Depression**



# **Fig 2. Graphical representation of Canopy Temperature Depression**

# **Corelation between Canopy Temperature, Canopy Temperature Depression, Grain Yield and Biological Yield.**

The correlation analysis as shown in Table 4.7 exhibits a positive correlation of grain yield and biological yield with CTD at 68 DAS indicating cooler canopy temperature leading to more grain yield and increase in biomass. Definitely, cooler canopy is an indicator of good plant water status and functions. Similar inference was provided by Karimizadeh *et al.,* 2011, Talebi *et al.,* 2011 and Guendouz *et al.,* 2012, Mondal *et al*., 2013. Thus, CTD is more important parameter in selecting tolerant genotypes as concluded by Blum *et al.,* 1988 and Balota *et al.,* 2007.

<b>Characters</b>	$CT_1$	CT <sub>2</sub>	$\mathbf{CTD}_1$	$\mathbf{CTD}_2$	GY	$\mathbf{B}\mathbf{Y}$
$CT_1$	1					
CT <sub>2</sub>	$-0.012$					
$CTD_1$	$-0.529$ **	$-0.014$	1			
$\mathbf{CTD}_2$	0.119	$-0.738**$	0.021			
<b>GY</b>	$-0.004$	0.008	$0.289**$	$-0.035$		
BY	$-0.109$	$-0.026$	$0.362***$	0.029	$0.477***$	

**Table 4.7.: Correlation Matrix between CT, CTD, GY and BY**

**\***significant at 5% probability level, **\*\***significant at 1% probability level, **CT1**= Canopy Temperature at 68 DAS, **CT2**= Canopy Temperature at 93 DAS, **CTD1**= Canopy Temperature Depression at 68 DAS, **CTD2**= Canopy Temperature Depression at 93 DAS, **GY**= Grain Yield (t-ha) and **BY**= Biological Yield (t-ha).

# **4.3. Study on Spot blotch) resistance**

Spot blotch (*Bipolaris sorokiniana* (Sacc.) Shoem) scoring was done at four crop growth stages in double-digit scale (00-99) developed by Saari and Prescott, 1975. Disease severity and AUDPC (Area Under Disease Progress Curve) was calculated as per Wilcoxson *et al.,* (1975).

Analysis of variance clearly revealed highly significant effect of genotype as well as growth stages on disease severity % (Table 4.8a). Interaction between genotype and growth stages was not found significant. Mean values of severity among different growth stages indicated gradual increase in severity along with increase in growth stages (Table 4.8c). This is obvious in spot blotch resistance where disease progress rapidly with the advances in maturity of crop especially in susceptible genotypes (Joshi *et al.,* 2007; Duveiller, 2005).

**Table 4.8a.: Two-way Analysis of variance for Disease severity (%) under different growth stages**

<b>Source of variation</b>	df	Mean sum of square	F pr.
Genotype	49	405.65***	< 0.001
Growth stage	3	85734.72***	< 0.001
Genotype x Growth stage	147	66.88	0.301
Error	199	61.81	

**Table 4.8b. : Statistical parameters for Disease severity (%)** 



<b>GENOTYPE</b>	<b>88 DAS</b>	<b>95 DAS</b>	<b>102 DAS</b>	<b>109 DAS</b>
1	19.753	28.951	50.556	63.889
$\mathbf 2$	25.185	42.963	72.901	97.222
3	36.790	41.049	69.877	88.333
4	35.123	42.469	67.901	95.556
5	29.630	44.938	67.099	96.111
6	28.704	42.160	81.790	96.667
$\overline{7}$	34.753	38.025	62.531	95.000
8	19.691	33.704	77.284	94.444
$\boldsymbol{9}$	18.086	28.889	61.790	88.333
10	27.778	47.531	71.481	92.778
11	19.321	34.506	71.358	93.889
12	19.691	27.407	58.765	86.111
13	24.383	41.358	56.358	94.444
14	34.198	40.864	58.704	87.778
15	26.235	31.728	76.667 73.889	95.556
16	32.003 31.790	48.951 48.889		92.222 93.889
17 18	34.877	42.840	91.728 63.210	93.333
19	32.099	47.531	85.494	96.667
20	28.333	45.370	85.000	97.222
	25.988		60.432	77.222
21		24.074 54.444	83.889	97.222
22	35.494			
23	26.235	35.309	71.605	95.556
24	22.901	19.753	62.531	88.889
25	27.716	29.198	77.963	88.889
26	38.642	41.605	77.346	93.333
27	26.914	39.383	71.667	94.444
28	22.222	32.037	70.864	88.333
29	21.296	28.148	51.667	78.333
30	30.741	48.889	88.333	99.444
31	30.309 33.395	56.790 49.506	79.444 81.420	96.667 95.000
32	25.556	47.901	73.333	
33				97.222
34	33.457	41.790	81.111	95.556
35	40.247	57.284	95.556	99.444
36 37	21.420 36.728	26.358 48.148	75.617 91.111	85.000 98.889
38				
	44.259	49.630	81.667	95.000
39	19.938	30.617 41.235	56.481	79.444
40	27.716	43.086	90.000	98.333
41	36.728 37.840	41.790	78.704 80.247	93.333 93.889
42 43	24.012	44.444	98.889	100.000
44	27.160	41.975	61.235	84.444
45	30.926	37.346	70.741	92.778
46	33.889	46.173	84.444	98.333
47	25.617	34.383	73.889	90.000
48	33.889	52.037	71.111	96.667
49	35.432	44.630	88.765	95.000
50	30.247	59.136	93.025	99.444
<b>Mean</b>	29.307	40.944	74.549	92.511

**Table 4.8c. : Two-way mean table for Disease severity (%) under different growth stages**

To identify the progress of disease along with growth stages AUDPC values was calculated for each genotype. Table 4.9 shows the mean values of AUDPC. In AUDPC 1 values the maximum value is shown by ENTRY 35 (341.36) and the minimum value is shown by ENTRY 24 (149.29). In AUDPC 2 the maximum value is exhibited by ENTRY 35 (534.94) whereas minimum by ENTRY 1 (278.27). In AUDPC 3 the maximum value is shown by ENTRY 43 (696.11) whereas minimum by ENTRY 1 (400.555). In the Mean AUDPC values the maximum is shown by ENTRY 35 (519.595) whereas the minimum value by ENTRTY 1 (283.095).

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 Liatukas and Ruzgas, 2012. Among the genotypes 29 were highly susceptible (HS), 14 were susceptible to highly susceptible(S-HS) and 06 were susceptible(S). Only one genotype i.e., ENTRY 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 were either highly susceptible or susceptible category. Thus, no genotype could be selected in terms of disease resistance parameter.

Genotype	<b>AUDPC1</b>	<b>AUDPC 2</b>	<b>AUDPC 3</b>	<b>Mean AUDPC</b>	<b>Resistance category</b>
1	170.465	278.270	400.555	283.095	$MS-S$
$\overline{2}$	238.515	405.525	595.430	413.160	$\rm{HS}$
3	272.440	388.240	553.735	404.805	HS
$\overline{\mathbf{4}}$	271.575	386.300	572.100	409.990	HS
$\overline{\mathbf{5}}$	260.985	392.130	571.235	408.115	HS
6	248.025	433.825	624.595	435.485	HS
$\overline{7}$	254.720	351.945	551.355	386.010	S-HS
8	186.885	388.455	601.050	392.130	S-HS
9	164.415	317.380	525.430	335.740	${\bf S}$
10	263.580	416.545	574.905	418.340	$\overline{HS}$
11	188.395	370.525	578.360	379.095	S-HS
12	164.845	301.605	507.065	324.510	$\overline{S}$
13	230.090	342.005	527.810	366.635	S-HS
14	262.715	348.485	512.685	374.630	S-HS
$\overline{15}$	202.870	379.380	602.780	395.010	S-HS
16	283.335	429.940	581.390	431.555	HS
17	282.375	492.160	649.660	474.735	HS
18	272.005	371.170	547.905	397.025	S-HS
$\overline{19}$	278.705	465.585	637.560	460.615	$H\overline{S}$
20	257.965	456.295	637.780	450.680	HS
$\overline{21}$	175.215	295.770	481.790	317.590	$\overline{S}$
$\overline{22}$	314.785	484.170	633.890	477.615	$\overline{HS}$
$\overline{23}$	215.405	374.200	585.060	391.555	S-HS
24	149.290	287.995	529.970	322.415	S
$\overline{25}$	199.195	375.060	583.985	386.080	$S-HS$
26	280.865	416.330	597.375	431.525	HS
$\overline{27}$	232.035	388.670	581.385	400.700	HS
28	189.905	360.155	557.190	369.085	S-HS
29	173.055	279.350	455.000	302.470	S
30	278.700	480.275	657.220	472.065	HS
31	304.845	476.820	616.390	466.020	<b>HS</b>
32	290.155	458.240	617.465	455.290	HS
33	257.095	424.320	596.945	426.120	HS
34	263.365	430.150	618.335	437.285	HS
$\overline{35}$	341.360	534.940	682.500	519.595	HS
$\overline{36}$	167.220	356.915	562.160	362.100	$S-HS$
37	297.070	487.405	665.000	483.155	HS
38	328.610	459.535	618.335	468.830	HS
39	176.945	304.850	475.740	319.175	S
40	241.325	459.320	659.170	453.270	<b>HS</b>
41	279.350	426.265	602.130	435.915	HS
42	278.705	427.130	609.475	438.440	HS
43	239.600	501.665	696.110	479.125	HS
44	241.975	361.235	509.875	371.030	S-HS
45	238.950	378.300	572.315	396.525	S-HS
46	280.215	457.160	639.720	459.035	<b>HS</b>
47	210.000	378.950	573.610	387.520	S-HS
48	300.740	431.020	587.225	439.660	<b>HS</b>
49	280.215	466.885	643.180	463.425	<b>HS</b>
50	312.840	532.565	673.645	506.345	HS
$S.Em (\pm)$	26.634	38.896	35.310	29.865	
L.S.D. (0.05)	75.693	110.541	100.349	84.871	

**Table 4.9. : AUDPC values pertaining to different genotypes under study**

# **4.4. Genetic divergence analysis (D<sup>2</sup> analysis)**

Chi-square test indicated that there was sufficient divergence in the genotypes and hence  $D^2$  analysis (Mahalanobis, P.C., 1936) was done. The distribution of genotypes into 5 different clusters was made on the basis of  $D^2$  analysis based on 15 characters namely GER= Germination per meter, DF= 50% Heading, PH= Plant Height (cm), AL= Awn Length (cm), SL= Spike Length (cm), GPS= Grain per Spike, SPS= Spikelet per spike, TM= Tiller per Meter, AUCIPC= area under chlorophyll index progress curve, CTD= Canopy Temperature Depression, AUDPC=area under disease progression curve, TGW= 1000 Grain Weight (g), BM= Biological yield ( $t$ <sup>-ha</sup>), GY= Grain Yield ( $t$ <sup>-ha</sup>), and HI= Harvest Index. Highest no. of genotypes (17) was accommodated in Cluster I and II (Table 4.10) while cluster III contained 12 genotypes. Smallest cluster was cluster IV with only single genotype while Cluster V had 3 only.

The average intra (diagonal) and inter (off-diagonal) cluster  $D^2$  were presented in Table 4.11. The inter-cluster distances were found higher than the intra-cluster distances which indicated the wider genetic diversity among the tested genotypes of different groups. Similar findings were reported by Samal and Jagadev (1996), Ahmed *et al.* (2002) and Zaman *et al,* 2014. Maximum inter cluster  $D^2$  was exhibited by the Cluster I and V (1243.168) closely followed by Cluster I and IV (1138.777). The other inter cluster differences such as Cluster I and III (877.550), Cluster II and IV (472.026) and Cluster II and V (420.085) was found moderate to low. High intra cluster distance was exhibited by cluster II (101.703) closely followed by Cluster I (101.444) as they contained highest number of genotypes. From the intra and inter cluster  $D^2$  values, it was revealed that the genotypes from the most divergent clusters like I and V could be selected as parents for hybridisation to get wide spectrum of variation within the segregating material.

<b>Cluster No.</b>	Number of genotypes	Name of genotypes
	17	ENTRY 1, ENTRY 2, ENTRY 3, ENTRY 4, ENTRY 5, ENTRY 6, ENTRY
		7, ENTRY 8, ENTRY 9, ENTRY 10, ENTRY 11, ENTRY 12, ENTRY 13,
		ENTRY 14, ENTRY 15, ENTRY 16, ENTRY 17
$\mathbf H$	17	ENTRY 18, ENTRY 19, ENTRY 20, ENTRY 21, ENTRY 22, ENTRY 23,
		ENTRY 24, ENTRY 25, ENTRY 26, ENTRY 27, ENTRY 28, ENTRY 29,
		ENTRY 30, ENTRY 31, ENTRY 33, ENTRY 34, ENTRY 35
$\Pi$	12 <sub>1</sub>	ENTRY 32, ENTRY 36, ENTRY 38, ENTRY 39, ENTRY 40, ENTRY 41,
		ENTRY 42, ENTRY 43, ENTRY 44, ENTRY 45, ENTRY 47, ENTRY 48
$\bf{IV}$		<b>ENTRY 37</b>
	3	ENTRY 46, ENTRY 49, ENTRY 50

**Table 4.10. : Distribution of 50 wheat genotypes into different clusters.**

<b>Cluster No.</b>		$\mathbf{I}$	III	IV	V
	101.444	343.788	877.550	1138.777	1243.168
$\mathbf{I}$		101.703	245.427	472.026	420.085
Ш			74.069	226.630	119.553
IV				$\boldsymbol{0}$	322.125
					75.990

**Table 4.11. : Average intra (diagonal) and inter (off diagonal) cluster D<sup>2</sup> values of 50 genotypes of wheat**

The cluster mean vales for 15 characters along with their contribution to divergence have been given in Table 4.12. Highest grain yield as well as biological yield was produced by cluster I (3.399 and 9.277 ton –ha. respectively). At the same time, highest chlorophyll retention value was exhibited by cluster I having highest AUCIPC value (2345.88). In terms of disease infestation, it was also found that lowest AUDPC value had been shown by cluster I (390.179) which indicated less disease infestation for this cluster. Thus, it could be concluded that most promising genotypes in terms of yield, physiological efficiency and disease susceptibility have been grouped in cluster I.

The maximum contribution to the genetic divergence was exhibited by the character CTD (80.571%), followed by HI (10.286%), GER (8.163%) and GPS (8.163%). Highest CTD value was shown by Cluster V (12.712) which indicated that promising genotypes for this trait had been accommodated in this cluster.



# **Table 4.12. : Cluster mean for 15 characters of wheat genotypes**

**GER**= Germination per meter, **DF**= 50% Heading, **PH**= Plant Height (cm), **AL**= Awn Length (cm), **SL**= Spike Length (cm), **GPS**= Grain Per Spike, **SPS**= Spikelet Per Spike, **TM**= Tiller per Meter, **TGW**= 1000 Grain Weight (g), **GY**= Grain Yield (t<sup>-ha</sup>), **BY**= Biological Yield (t<sup>-ha</sup>), **AUCIPC**= Area Under Chlorophyll Index Progress Curve, **CTD**= Canopy Temperature Depression, **AUDPC**= Area Under Disease Progression Curve, **HI**= Harvest Index

# **4.5. 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  $D<sup>2</sup>$  analysis. Among the fifteen principal components (PCs) only five components (PCs) had showed Eigen value >1.00, which accounted for 69.03% of cumulative proportion of variance (Table 4.13). Among them PC 1 and PC 2 accounted for 26.01% and 13.08% variance respectively.

PC	<b>Eigen value</b>	% Variance
$\mathbf{1}$	3.90	26.01
$\overline{2}$	1.96	13.08
3	1.84	12.26
$\overline{\mathbf{4}}$	1.61	10.71
5	1.05	6.97
6	0.98	6.54
$\overline{7}$	0.81	5.39
8	0.70	4.68
9	0.53	3.52
10	0.51	3.41
11	0.36	2.37
12	0.30	2.03
13	0.25	1.65
14	0.19	1.27
15	0.02	0.12

**Table 4.13. : Summary of the contribution of the different principal components to variability**

**Fig 3: Scree plot analysis for PCA**



Loading value of different characters are presented in Table 4.14. 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 positively and negatively contributed more to the diversity and they were the ones that were responsible for creating difference between clusters. In PC 1, characters such as BY, PH, GPS, AUCIPC had high positive loadings and thus contributed positively while AUDPC and CTD had negative loadings which contributed negatively. In PC 2, GY and HI had high positive loadings while DF had negative loading. The findings of PCA revealed that these effective contributing traits in PC 1 and PC 2 had the significant role in diversification of genotypes and selection may be possible based on this trait for future breeding programmes.

<b>Characters</b>	PC <sub>1</sub>	PC <sub>2</sub>	PC <sub>3</sub>	PC <sub>4</sub>	PC <sub>5</sub>
<b>GER</b>	0.2044	0.1674	0.4115	0.3483	0.1114
DF	0.1483	$-0.4193$	$-0.1469$	0.2245	0.0209
PH	0.3916	$-0.1670$	$-0.1830$	$-0.0124$	0.1806
AL	0.1347	0.1509	$-0.5108$	0.2437	0.0162
SL	0.1982	0.1738	$-0.1084$	$-0.4096$	0.1561
<b>GPS</b>	0.3066	0.0473	0.3282	$-0.1541$	0.0201
<b>SPS</b>	0.2640	$-0.0695$	0.4692	$-0.2545$	0.1169
TM	$-0.0038$	0.0833	0.2169	0.6022	$-0.1689$
<b>TGW</b>	0.0629	$-0.1999$	0.1255	$-0.2352$	$-0.6361$
GY	0.2695	0.5402	$-0.0723$	0.0612	0.0481
BY	0.4469	0.0284	$-0.0060$	0.1480	0.1758
<b>AUCIPC</b>	0.3043	$-0.0897$	$-0.2653$	$-0.1880$	0.0271
<b>CTD</b>	$-0.2739$	$-0.2293$	$-0.0084$	0.0197	0.4909
<b>AUDPC</b>	$-0.2804$	0.1092	0.1714	$-0.1157$	0.4324
$\mathbf{H}$	$-0.1981$	0.5442	$-0.0849$	$-0.1403$	$-0.1441$

**Table 4.14. : Five principal components along with their factor loadings**

**GER**= Germination per meter, **DF**= 50% Heading, **PH**= Plant Height (cm), **AL**= Awn Length (cm), **SL**= Spike Length (cm), **GPS**= Grain Per Spike, **SPS**= Spikelet Per Spike, **TM**= Tiller per Meter, **TGW**= 1000 Grain Weight (g), **GY**= Grain Yield (t-ha), **BY**= Biological Yield (t-ha), **AUCIPC**= Area Under Chlorophyll Index Progress Curve, **CTD**= Canopy Temperature Depression, **AUDPC**= Area Under Disease Progression Curve, **HI**= Harvest Index

PCA biplot graph (Fig 4) also showed the divergence of the present wheat population with respect to 15 quantitative traits. Strong positive correlation was found between GY, BY, GPS while negative association between GY and DF , CTD and PH. These findings would be helpful for making selection criteria of individual genotypes under present environmental situation.

**Fig 4. PCA Biplot graph showing 50 genotypes by points (G-1 to G-50) and trait vectors as green arrow**



# **4.6 Hierarchical clustering of 50 genotypes based on Euclidean distance**

Hierarchical clustering of 50 genotypes of wheat was done based on Euclidean distance (Fig 5.). It revealed 5 clusters as shown in Table 4.15.





If we compare the both clusters (Table 4.10 and Table 4.15), we found lot of dissimilarity between them. Clustering based on Euclidean distance, grouped the 50 genotypes

into 5 Clusters within which Cluster I had the most genotypes (18), followed by Cluster IV (17). Moreover, Cluster II, III, and V comprised of 8, 5, and 2 genotypes respectively. The probable reason for this dissimilarity is that the Mahalanobis distance is a metric that takes into account both inter-variable correlations and variance differences. Variables with high variation and strongly linked variables are given less weight by the Mahalanobis distance, ensuring that all qualities are treated equally (Mimmack *et al.,* 2001). Whereas the Euclidean distance assigns equal weight to each variable and it gives excess weight to correlated variables (Jolliffe 1986).



# **Fig 5. Hierarchical clustering of 50 genotypes of wheat based on Euclidean distance**

# **4.7. Correlation analysis**

# **Correlation between 15 characters of wheat**

The correlation analysis (Table 4.16) revealed that among the 15 characters only eight of them were positively associated with grain yield. Those traits included GER, PH, AL, SL, GPS, AUCIPC, BY and HI. GER was also positively correlated with GPS, SPS, TM, BY and GY. DF was positively correlated with PH and negatively correlated with AUDPC and HI. PH was positively correlated with DF, AL, BY and GY, whereas, negatively correlated with HI. AL was positively correlated with PH, CTD, BY and GY, whereas negatively correlated with SPS. SL was positively correlated with GY only. GPS is positively correlated with GER, SPS, BY and GY. SPS was positively correlated with GER, GPS and BY whereas negatively correlated with AL only. TM was positively correlated with GER only.

Among the physiological traits, AUCIPC was found positively correlated with PH, CTD, BY and GY. Similarly, CTD was positively correlated with AUCIPC and BY. This indicated that with higher AUCIPC value the biomass of the genotypes also increased as well as canopy temperature decreased due to high physiological efficiency. Similar result was found in wheat Khakwani *et al.,* 2012 and Abdul *et al.,* 2018.

AUDPC was found to be positively correlated with HI whereas negatively correlated with DF, PH, GPS, AUCIPC and BY. This indicated that higher disease severity was negatively associated with high chlorophyll index value. This might be due to loss of greenness during high disease infestation. Similar finding was found by Rosyara *et al.,* 2007, Rosyara *et al.,* 2010.

<b>Characters</b>	<b>GER</b>	DF	PH	AL	${\bf SL}$	<b>GPS</b>	<b>SPS</b>	TM	<b>AUCIPC</b>	<b>CTD</b>	<b>AUDPC</b>	<b>TGW</b>	BY	GY	$\mathbf{H}$
<b>GER</b>	$\mathbf{1}$														
DF	$-0.044$	1													
PH	0.124	$0.218*$													
AL	0.008	0.096	$0.236*$	$\mathbf{1}$											
SL	$-0.055$	$-0.038$	0.118	0.069	$\mathbf{1}$										
<b>GPS</b>	$0.396**$	0.122	0.115	0.010	0.190	$\mathbf{1}$									
<b>SPS</b>	$0.356**$	0.107	0.130	$-0.262**$	0.178	$0.642**$	1								
TM	$0.403***$	0.122	$-0.152$	0.046	$-0.124$	0.123	0.109	$\mathbf{1}$							
<b>AUCIPC</b>	0.116	0.046	$0.442**$	0.179	0.190	0.187	0.083	$-0.186$	1						
<b>CTD</b>	$-0.017$	0.100	$0.312**$	$0.220*$	$-0.009$	0.004	0.023	0.001	$0.294***$	1					
<b>AUDPC</b>	$-0.088$	$-0.309**$	$-0.394**$	$-0.156$	$-0.053$	$-0.199*$	$-0.104$	0.023	$-0.262**$	$-0.021$	1				
<b>TGW</b>	0.058	0.002	$-0.151$	$-0.111$	0.079	0.151	0.189	0.133	0.040	$-0.074$	0.029	$\mathbf{1}$			
BY	$0.328***$	0.115	$0.723***$	$0.249*$	0.156	$0.357**$	$0.267**$	0.011	$0.402**$	$0.239*$	$-0.382**$	$-0.165$	$\mathbf{1}$		
<b>GY</b>	$0.276**$	$-0.136$	$0.286**$	$0.240*$	$0.316**$	$0.230*$	0.049	0.090	$0.217*$	0.144	$-0.166$	$-0.103$	$0.477**$	$\mathbf{1}$	
H <sub>I</sub>	$-0.095$	$-0.258**$	$-0.394**$	0.006	0.169	$-0.088$	$-0.170$	0.052	$-0.173$	$-0.090$	$0.202*$	0.053	$-0.473**$	$0.515***$	$\mathbf{1}$

**Table 4.16. : Correlation Matrix between 15 characteristics of wheat.**

**\***significant at 5% probability level, **\*\***significant at 1% probability level.

**GER**= Germination per meter, **DF**= 50% Heading, **PH**= Plant Height (cm), **AL**= Awn Length (cm), **SL**= Spike Length (cm), **GPS**= Grain Per Spike, **SPS**= Spikelet Per Spike, **TM**= Tiller per Meter, **AUCIPC**= Area Under Chlorophyll Index Progress Curve, **CTD**= Canopy Temperature Depression, **AUDPC**=Area Under Disease Progression Curve, **TGW**= 1000 Grain Weight (g), **BM**= Biological Yield (t-ha), **GY**= Grain Yield (t-ha), **HI**= Harvest Index
# *Chapter 5 Summary and Conclusion*

The ANOVA for the 12 characters indicated significant differences between the genotypes for 50% Heading, Plant height, Awn length, Spike length, Spikelet per spike, 1000 grain weight and biological yield.

Mean germination per meter was 44.05 which ranged between 33.5-65 and had a moderate CV (19.75%) and poor heritability (0.089) and genetic advance (3.78%). Range of days to flowering varied between 58 to 78 days with a mean value of 66.62 and it had low CV (4.71%) and high heritability (0.645) and genetic advance (10.5%) is moderate for this trait. Plant height varied between 79.20 cm to 98.30 cm with a mean value of 86.62 cm and low CV (4.98%) and moderate heritability (0.354) and poor genetic advance (4.52%). Mean awn length was 7.12 cm and it varied between 1.80 cm-8.90 cm with low CV (6.68%) and high heritability (0.792), genetic advance (23.95%). The mean of spike length was 10.17 cm which ranged between 8.50 cm-17.70 cm with high CV (15.02%) and low heritability (0.233). The grain per spike had a mean of 48.50 which ranged between 36.00-61.50 with moderate CV (13.47%) and low heritability (0.177). The mean of spikelet per spike was 18.61 which ranged between 15.40-21.60 with low CV (6.43%) and moderate heritability (0.406) and low genetic advance (6.98%). Mean tiller per meter was 61.18 with a range of 47.50-76.34 with moderate CV (13.24%) and very low heritability (0.092) and genetic advance (2.69%). Mean 1000 grain weight was 39.71g and it ranged between 32.27g-46.81g with low CV (7.23%) and moderate heritability (0.490) and genetic advance (10.24%). Mean grain yield was  $3.24$  t<sup>-ha</sup> and it ranged between 2.3-4.3  $t^{-ha}$  with a high CV (20.38%) and poor heritability (0.044). However, mean biological yield was 8.76 t<sup>-ha</sup> and it ranged between 6.33-12.77 t<sup>-ha</sup> and heritability (0.309) and genetic advance (12.08%) is moderate for the trait. Harvest index ranged between 0.27-0.55 with a mean value of 0.38, the CV of this trait was high (21.78%) whereas heritability was very poor (0.077).

Among the physiological traits Chlorophyll Index (CI) studied at four different crop growth stages (88 DAS, 95 DAS, 102 DAS and 109 DAS) showed significant difference among the genotypes as well as growth stages. The mean value of CI indicated gradual decline of chlorophyll index with advances in growth stages. However, the rate of decrease in CI value was found highly variable among the 50 genotypes under study. Area Under Chlorophyll Index

Progress Curve (AUCIPC) showed highest value in genotype ENTRY 1 which indicated higher retention of chlorophyll at maturity in this genotype.

Canopy Temperature Depression studied at two different growth stages (68 DAS and 93 DAS) showed significant difference among the growth stages only. 13 genotypes (ENTRY 5, 8, 14, 18, 20, 21, 24, 28, 34, 35, 46, 48 and 49) showed higher CTD value at later stages which indicating high physiological efficiency for these genotypes. Correlation study between CT, CTD, GY and BY revealed positive association of grain yield and biological yield with CTD value at 68 DAS indicating cooler canopy temperature leading to higher grain yield and increased biomass. Definitely, cooler canopy is an indicator of good plant water status and functions. Similar results were shown by Karimizadeh *et al.,* 2011, Talebi *et al.,* 2011 and Guendouz *et al.,* 2012.

Spot blotch (*Bipolaris sorokiniana* (Sacc.) Shoem) scoring was done at four crop growth stages in double-digit scale (00-99). Disease severity and AUDPC (Area Under Disease Progress Curve) was calculated. Analysis of variance clearly revealed highly significant effect of genotype as well as growth stages on disease severity %. Mean disease severity under different growth stages indicated gradual increase in severity with the advancement in maturity. This is obvious in spot blotch resistance where disease progress rapidly with the advances in maturity of crop especially in susceptible genotypes (Joshi *et al.,* 2007).

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 (Liatukas and Ruzgas, 2012). The grouping separates the 50 genotypes into 6 as susceptible, 14 as susceptible to highly susceptible and 29 as highly susceptible. The only moderately susceptible to susceptible is ENTRY 1. No genotype was found either moderately resistant (MR) or resistant (R) category.

The 50 genotypes were classified into five clusters as per  $D^2$  analysis. The distribution of genotypes into 5 different clusters on the basis of  $D^2$  analysis based on 15 characters indicated substantial genetic diversity. The maximum inter cluster  $D^2$  value was exhibited by the Cluster I and V (1243.168) closely followed by Cluster I and IV (1138.777). The maximum intra cluster  $D^2$  was exhibited by Cluster II (101.703) closely followed by Cluster I (101.444). From this intra and inter cluster  $D^2$  value analysis it indicated that the genotypes from the most divergent Clusters viz. I and V could be selected as parents for hybridisation to get wide spectrum of variation. Maximum contribution to the genetic divergence was attributed by the character CTD, followed by HI, GER and GPS.

PCA was done by using all the fifteen characters that were used for  $D^2$  analysis. Among the fifteen principal components (PCs) only five components (PCs) had showed Eigen value >1.00, which accounted for 69.03% of cumulative proportion of variance. Biplot curve showed, in PC 1 characters such as BY, PH, GPS, AUCIPC had high positive loadings and thus contributed positively while AUDPC and CTD had negative loadings which contributed negatively. In PC 2, GY and HI had high positive loadings while DF had negative loading. Also, strong positive correlation was found between GY, BY, GPS while negative association between GY and DF , CTD and PH. These findings led to the conclusion that these effective contributing traits in PC 1 and PC 2 had the significant role in diversification of genotypes and selection might be possible based on this trait for future breeding programmes. Hierarchical clustering of 50 genotypes of wheat based on Euclidean distance exhibited dissimilarity with clustering using Mahalanobis distance due to difference in weightage provided to correlated variables by both the methods.

The correlation analysis among the 15 characters showed that only eight of them were positively associated with grain yield. Those traits included GER, PH, AL, SL, GPS, AUCIPC, BY and HI. Among the physiological traits, AUCIPC was found positively correlated with PH, CTD, BY and GY. Similarly, CTD was positively correlated with AUCIPC and BY. This indicated that with higher AUCIPC value the biomass of the genotypes also increased as well as canopy temperature decreased due to high physiological efficiency. Similar result was found in wheat Khakwani *et al.,* 2012 and Abdul *et al.,* 2018.

AUDPC was found to be positively correlated with HI whereas negatively correlated with DF, PH, GPS, AUCIPC and BY. This indicated that higher disease severity was negatively associated with high chlorophyll index value. This might be due to loss of greenness during high disease infestation. Similar finding was found by Rosyara *et al.,* 2007, Rosyara *et al.,* 2010.



**Plate No 1: Germination per meter calculation.**



**index using chlorophyll meter** 



**Plate No 3: Infrared Thermometer to measure Canopy Temperature.**



**Plate No 4: 19 HTWYT nursery at 100% heading stage**



**Plate No 5: 19 HTWYT nursery prior maturity stage.**



**Plate No 6: Spot Blotch in lower leaves of wheat.**

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## **ANNEXTURE**

## **A. Table of Chlorophyll Index at four growth stages and Area Under Chlorophyll Index Progress Curve (AUCIPC)**







#### **GEN REP CTD1 CTD2 M CTD 1** 2.54 -1.32 0.61 **1 2 1.18 1.18 0.88 1.03 1 1** 0.98 **0** 0.49 **2** 2.52  $-0.24$  1.14 **1 1 1.22 1.22 1.26 1.28 1.28 1.28 1.28 2 1.06** 1.66 **1.36 1 1 1.74 0.46 1.1 2** -0.14 0.42 0.14 **1 1 1 0.94 0.96 0.95 2** -0.24 1.08 0.42 **1 1 1.28 1.28 1.28 1.28 1.28 1.28 1.28 1.28 1.28 1.28 1.28 2 2 2 2.86 1.31 1** -0.14 0.76 0.31 **2** 2.26 0.86 1.56 **1 1 1 1.11 1.11 1.46 1.11 2 2 1 2.05 2.05 1 1 1.44 1.44 1.42 1.42 0.01 2** 2.72 1.68 2.2 **1** 2.32 -1.7 0.31 **2 1.04 0.3 1.067 1** 2.6 -0.7 0.95 **2 1.5 1.9 1.7 1** 0.84 -1.06 -0.11 **2** 0.44 1.34 0.89 **1 1 1 1.8 1.8 0.12 1.8 0.12 2 1.86 1.86 0.6 1.23 1 1 1 1 1.73 2** 0.04 1.58 0.81 **1 1 1 1 2.18 1 2.18 1 1 1 1 1 1 1 2** 1.92 0.12 1.02 **1 1 1.56 1.2 1.38 2** 0.76 0.08 0.42

#### **B. Table of Canopy Temperature Depression (CTD) at two growth stages.**





<b>GEN</b>	<b>REP</b>	DS% 1	DS% 2	DS% 3	<b>DS% 4</b>	<b>AUDPC1</b>	<b>AUDPC 2</b>	<b>AUDPC 3</b>	<b>MAUDPC</b>
$\mathbf{1}$	$\mathbf{1}$	17.40741	23.7037	48.2716	58.88889	143.8889	251.9136	375.0617	256.9547
$\mathbf{1}$	$\boldsymbol{2}$	22.09877	34.19753	52.83951	68.88889	197.037	304.6296	426.0494	309.2387
$\boldsymbol{2}$	$\mathbf{1}$	24.44444	32.22222	59.1358	95.55556	198.3333	319.7531	541.4198	353.1687
$\boldsymbol{2}$	$\boldsymbol{2}$	25.92593	53.7037	86.66667	98.88889	278.7037	491.2963	649.4444	473.1481
$\mathbf{3}$	$\mathbf{1}$	33.33333	30.74074	57.53086	81.11111	224.2593	308.9506	485.2469	339.4856
$\mathbf{3}$	$\boldsymbol{2}$	40.24691	51.35802	82.22222	95.55556	320.6173	467.5309	622.2222	470.1235
$\overline{\mathbf{4}}$	$\mathbf{1}$	33.7037	40.98765	57.65432	97.77778	261.4198	345.2469	544.0123	383.5597
4	$\boldsymbol{2}$	36.54321	43.95062	78.14815	93.33333	281.7284	427.3457	600.1852	436.4198
5	$\mathbf{1}$	23.08642	37.65432	48.76543	97.77778	212.5926	302.4691	512.9012	342.6543
5	$\boldsymbol{2}$	36.17284	52.22222	85.4321	94.44444	309.3827	481.7901	629.5679	473.5802
6	1	24.44444	33.95062	68.02469	93.33333	204.3827	356.9136	564.7531	375.3498
6	$\boldsymbol{2}$	32.96296	50.37037	95.55556	100	291.6667	510.7407	684.4444	495.6173
$\overline{7}$	$\mathbf{1}$	37.65432	35.4321	54.69136	96.66667	255.8025	315.4321	529.7531	366.9959
$\overline{7}$	$\overline{2}$	31.85185	40.61728	70.37037	93.33333	253.642	388.4568	572.963	405.0206
8	$\mathbf{1}$	14.07407	19.75309	57.90123	92.22222	118.3951	271.7901	525.4321	305.2058
8	$\boldsymbol{2}$	25.30864	47.65432	96.66667	96.66667	255.3704	505.1235	676.6667	479.0535
9	$\mathbf{1}$	18.14815	28.14815	51.11111	86.66667	162.037	277.4074	482.2222	307.2222
9	$\boldsymbol{2}$	18.02469	29.62963	72.46914	90	166.7901	357.3457	568.642	364.2593
10	$\mathbf{1}$	27.90123	42.83951	63.58025	90	247.5926	372.4691	537.5309	385.8642
10	$\boldsymbol{2}$	27.65432	52.22222	79.38272	95.55556	279.5679	460.6173	612.284	450.823
11	$\mathbf{1}$	23.08642	37.65432	64.44444	91.11111	212.5926	357.3457	544.4444	371.4609
11	$\boldsymbol{2}$	15.55556	31.35802	78.2716	96.66667	164.1975	383.7037	612.284	386.7284
12	$\mathbf{1}$	17.77778	24.19753	60	85.55556	146.9136	294.6914	509.4444	317.0165
12	$\boldsymbol{2}$	21.60494	30.61728	57.53086	86.66667	182.7778	308.5185	504.6914	331.9959

**C. Table of Disease Severity % at four growth stages with Area Under Disease Progress Curve (AUDPC).**

VII | P a g e







# **VITA**



## **EDUCATION QUALIFICATIONS**

# **Bachelor's degree:**



**Master's degree:**



# Curiginal

#### **Document Information**



#### Sources included in the report



1101 · https://www.thenharmainurnal.com/archives/2010/vol8issue2/DartK/8-2-06-633 ndf

 $1/37$ 

#### (SOURIK PODDAR) (DR. SAIKAT DAS)

Signature of student Chairman

Advisory committee