# Genetic Variability and Association Studies in Germplasm Accessions and Land Races of Wheat (Triticum aestivum L.)

#### **ABSTRACT**

Wheat (Triticum aestivum L.) is described as the 'King of Cereals' is a leading cereal crop which plays a crucial role in feeding the hungry world and improving global food security. Understanding the contribution of yield components to the variation of grain yield is essential for designing breeding programs and increasing grain production. A diversity panel of 200 genotypes including 145 accessions and 55 landraces of wheat were evaluated in Augmented Design for genetic variability and correlation between grain yield and yield related components. Analysis of variance revealed significant differences among the genotypes for all the traits except chlorophyll fluorescence. The highest phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) were recorded for effective number of tillers metre<sup>-1</sup> followed by biological yield row<sup>-1</sup>, flag leaf length, grain yield row<sup>-1</sup>. Heritability coupled with genetic advance percent of mean was high for effective number of tillers metre-1, followed by biological yield row<sup>-1</sup>, flag leaf length, grain yield row<sup>-1</sup>. Association studies revealed a highly significant and positive association of grain yield row<sup>-1</sup> with biological yield row<sup>-1</sup> (0.805), 1000grain weight (0.476), effective number of tillers metre-1 (0.328), flag leaf width (0.137) and harvest index (0.133) whereas, it showed a significant negative association with days to maturity (-0.304) and days to 50% heading, (-0.291). The study revealed that days to maturity, days to heading, 1000-grain weight and effective number of tillers metre-1 could be considered the most appropriate traits for improvement and selection of trait to achieve stable and high yielding early wheat genotypes.

**Keywords**: Correlation, Heritability, Genetic Advance, 1000-grain weight, Wheat.

## **INTRODUCTION**

Wheat (*Triticum aestivum* L.) having chromosome number, 2n= 6x=42 (AABBDD), belongs to the genus *Triticum* and family *Graminae* (*Poaceae*). Wheat is described as the 'King of Cereals' is a leading cereal crop which plays a crucial role in feeding the hungry world and improving global food security (Tiwari *et al.*, 2021). In comparison to all other food crops, this crop delivers greater nourishment to humans and is the second most popular food crop in the world and occupies a prominent position in Indian agriculture after rice. With the enormous increase in population by 2050, the world will require roughly 840 million tonnes of wheat, increasing from 764 million tonnes of today's production level. Similarly, wheat production in India must keep pace with population growth and by 2050, the country will require around 140 million tonnes of wheat. In terms of fulfilling the ever-

increasing domestic food demand, as well as generating foreign money, a considerable increase in production is essential (Sharma *et al.*, 2015). However, in the past recent years in wheat production yield plateau has been realized. Breeders face a challenge in increasing production since the rising population of the country will demand considerably more food than it does today. It is not possible to expand the production area. As a result, the only option left is to enhance productivity by developing superior varieties and improving agricultural production management to meet the growing food demand.

Landraces contain genes and gene complexes for quality traits, tolerance to biological and abiotic stresses, and adaptation to a wide range of low-input and organic farming systems (Jaradat, 2011). Thus, to improve yield and yield stability, especially under stress and variable climatic conditions, development of new varieties from landrace populations could be a viable strategy. The potential of wheat germplasm accessions and landraces are unexploited due to the lack of detailed information on the extent of adaptability and production status in the Bundelkhand region. The contribution of agromorphological traits of landraces of wheat towards grain yield enhancement under limited water conditions of the Bundelkhand region is still unknown. In this context, knowledge of the nature and degree of genetic variability is critical for the beginning of any systematic breeding effort (Islam *et al.*, 2004).

Grain yield is a complicated feature that is greatly impacted by a variety of genetic factors as well as environmental changes. In plant breeding programs, selecting solely for yield as such may be deceptive and that knowledge regarding genetic variability and the relationship between morphoagronomic characteristics and grain and yield is necessary for successful selection. The relationship between various features and grain yield is better understood through correlation studies (Dixet and Dubey, 1984). Studies on correlation coefficients between yield and yield components of various plant characters on yield and yield components have been reported **Pooja** et al., (2018), Rajput (2018), Kaur et al., (2019), Anubhav et al., (2020), Rai et al., (2020), Kumar et al., (2021) and Tsegaye et al., (2021).

In wheat, grain yield is a complex character which depends on its component traits. In any breeding population, the genetic improvement mainly depends upon the amount of genetic variability present. Thus, for genetic manipulation of grain yield and other characters in wheat, there is a need to examine the nature of genetic variability for the yield and related attributes. Estimation of genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) helps to choose the potential genotype, heritability assists in determining the response to selection and heritability coupled with genetic advance would be the most effective condition in predicting the resultant effect for selection of best genotypes for yield. Correlation studies enable to maximise crop yield by studying their interrelationship among various traits. Keeping this into consideration, the present investigation was

undertaken to better understand the genetic background and inheritance of the different traits in wheat genotypes. Furthermore, the specific objective of this study was to determine genetic variability, heritability, and correlations among the major quantitative traits of wheat in order to facilitate successful breeding programmes through efficient selection. The results of this study would aid in identifying the most relevant genetic material and plan the ensuing breeding programme to foster the varietal improvement initiatives.

#### MATERIALS AND METHODS

The experimental material comprised 145 accessions and 55 landraces of wheat genotypes was laid out at the Research Farm of Rani Lakshmi Bai Central Agricultural University, Jhansi, Uttar Pradesh (India) during *rabi* 2020-21. The research site was located at 25.51°N latitude and 78.54° E longitude and at an altitude of 284 metres above the mean sea level. The experiment was executed in augmented block design in 5 blocks with row to row spacing of 25 cm and plant to plant distance of 10 cm and standard agronomic practices were adopted. Observations were recorded for sixteen agro-morphological traits *viz.*, days to 50% heading (DF50), days to maturity (DM), flag leaf length (cm) (FLL), flag leaf width(cm) (FLW), awn length (cm) (AL), spike length (cm) (SL), peduncle length (cm) (PL), effective number of tillers metre-1(TPM), plant height (cm) (PH), leaf area index (LAI), chlorophyll fluorescence (CF), total chlorophyll content (CC), 1000-grain weight (g) (TGW), grain yield row<sup>-1</sup> (g) (GY), biological yield row<sup>-1</sup> (g) (BY) and harvest index (%) (HI). The chlorophyll content was measured by SPAD-502 chlorophyll meter as well as chlorophyll fluorescence was measured using LI-6400XT Portable Photosynthesis System(LI-COR, Nebraska USA)

The data were subjected to analysis of variance (Panse and Sukhatme, 1967) and further biometric procedures were applied to compute phenotypic and genotypic coefficient of Variation (GCV) (Burton, 1952), broad sense heritability (Burton and Devane, 1953) and genetic advance (Johnson *et al.* 1955), and character association (Singh and Chaudhary, 1979) were computed. Statistical analysis was carried out by R Programming.

#### **RESULTS AND DISCUSSION**

#### Variability Studies:

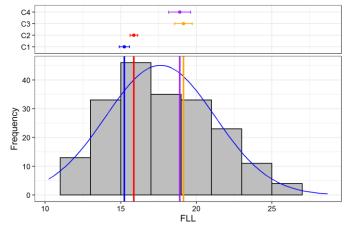
Analysis of variance demonstrated that the mean sum of squares for all checks and genotypes were highly significant except for chlorophyll fluorescence (**Table 1**). The frequency distribution of different quantitative traits in wheat are presented in **Fig. 1** 

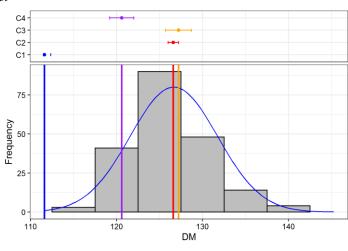
Table 1: Analysis of variance (ANOVA) for different yield-related traits in wheat

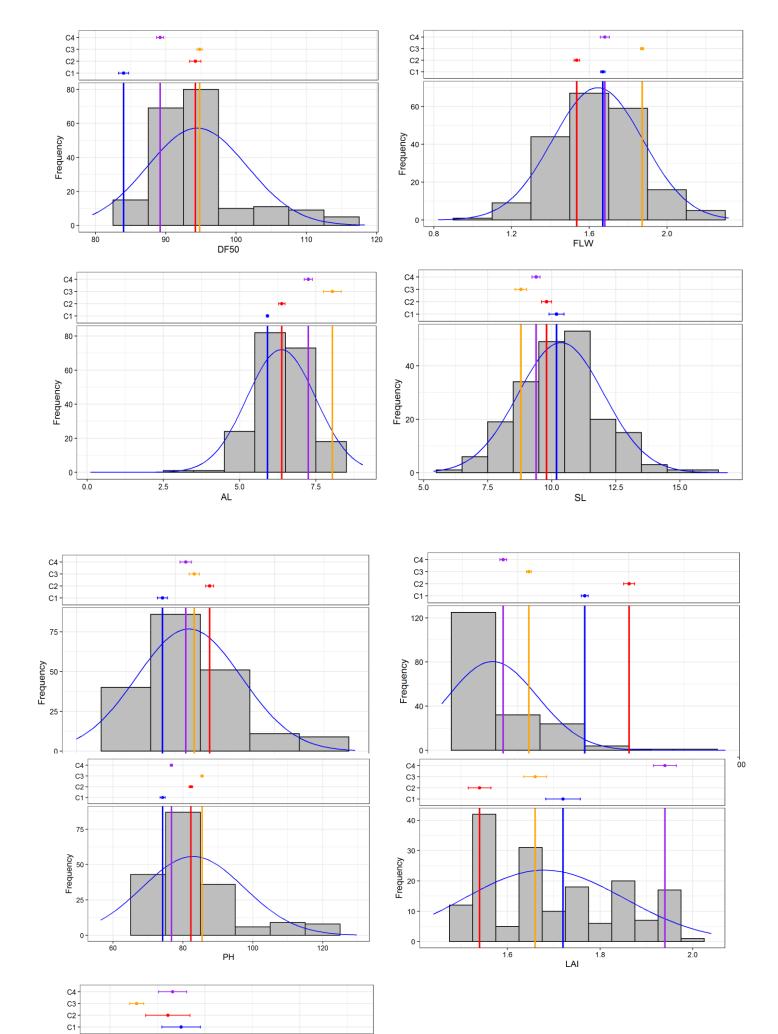
Source	Block	Entries	Check	Varieties	Checks vs.	Error		
	(adjusted)	(adjusted)			Varieties			
Df	4	203	3	199	1	12		
DF50	2.93	37.17 **	126.85 **	48.74 **	291.64 **	1.56		
DM	4.88	24.56 **	262.2 **	24.53 **	502.09 **	6.91		
FLL	1.05	12.44 **	20.45 **	12.77 **	2.03	1.37		
FLW	0.00044	0.05 **	0.1 **	0.06 **	0.04 **	0.0014		
AL	0.23	1.31 **	4.44 **	1.31 **	5.38 **	0.11		
SL	0.06	2.73 **	1.76 *	2.78 **	12.62 **	0.31		
PL	0.6	25.77 **	19.76 **	27.68 **	0.17	1.5		
TPM	20.3	512.87 **	3247.13 **	394.66 **	18253.44 **	13.13		
PH	1.39	184.41 **	132.46 **	214.76 **	195.83 **	1.21		
LAI	0.01	0.03 **	0.14 **	0.03 **	0.02 **	0.0026		
CF	0.00004	0.00016 *	0.000084	0.00017 *	0.00019	0.000069		
CC	1.2	7.54 **	7.11 *	8.05 **	21.73 **	1.71		
TGW	1.43	34.74 **	182.18 **	34.05 **	61.56 **	5.56		
GY	37.08	1931.81 **	3852.4 **	1843.43 **	29200.1 **	26.94		
BY	53.49	15292.43 **	46177.5 **	14680.49 **	86776.74 **	60.39		
HI	0.00016	0.0022 **	0.01 **	0.0022 **	0.01 **	0.000072		

DH –Days to 50% heading, DM-Days to maturity, FW-Flag leaf width, FL-Flag leaf length, AL-Awn length, SL-Spike length, PL-Peduncle length, TPM-Effective Tillers number per meter, PH-Plant height, LAF- leaf area index, CF-chlorophyll fluorescence, CC-chlorophyll content, TGW-1000- grain weight, GY-Grain yield per row, BY- Biological yield per row, HI-Harvest index.

\* Significant at 5 per cent level, \*\*Significant at 1 per cent level







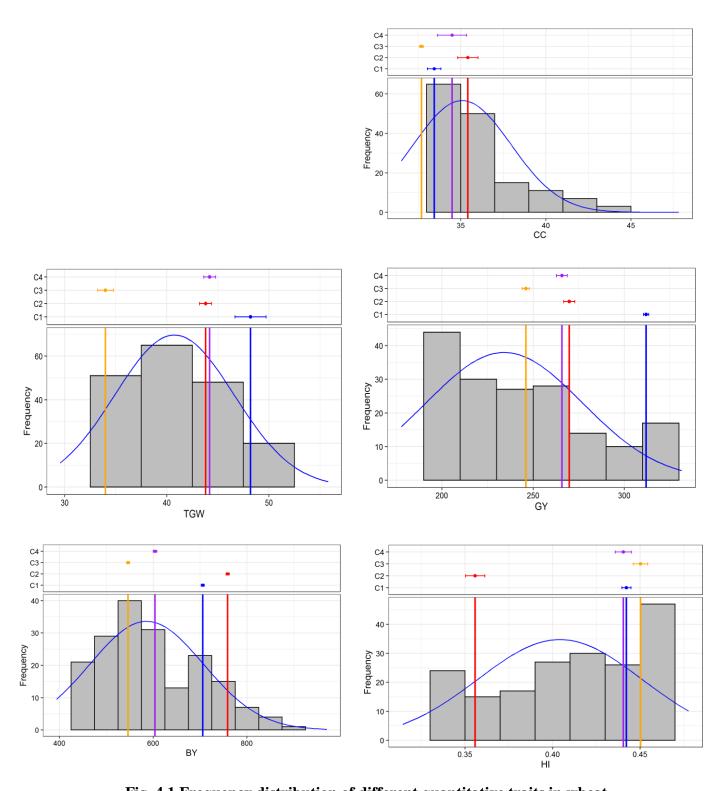


Fig. 4.1 Frequency distribution of different quantitative traits in wheat

This revealed that there was significant genetic variability for yield and yield-related parameters among the genotypes under study and it offers a strong chance of improving traits of interest in wheat breeding program. Kumar et al., (2013), Berhanu et al., (2017), Ahmed et al., (2018), Bhanu (2018), Wani et al., (2018), Kaur et al., (2019), Bayisa et al., (2020) and Sood et al., (2021), also came to similar conclusions and found that wheat genotypes differed in yield and yield-related characteristics. For each character under study, the phenotypic and genotypic coefficients of variation, broad sense heritability and expected genetic advance expressed as percentage of mean were evaluated and presented in Table

**2and**In the above graphs, C1, C2, C3, C4 indicate HD 2967, WB 2, UP 2903, HD 3086 and HD 3086 respectively.

In the present investigation, the highest GCV was observed for biological yield row<sup>-1</sup>, followed by effective number of tillers metre-1(Fig. 2). Similar findings were concluded by Wahidy et al., (2016) for biological yield row<sup>-1</sup> and Jan and Kashyap (2018) for effective number of tillers metre-1. Moderate GCV was reported for flag leaf length, grain yield row<sup>-1</sup>, plant height, awn length, peduncle length, spike length, flag leaf width, 1000- grain weight and harvest index. Similar conclusions were reported by Safi et al., (2017), Regmiet al., (2021), for peduncle length, Jan and Kashyap (2018) for flag leaf length, Safi et al., (2017), Alemu et al., (2019) and Kaur et al., (2019) for harvest index, Singh, A. G., & Sharma, A. K. (2021)forspike length and 1000-grain weight and Sood et al., (2021) for 1000-grain weight and harvest index. The lowest GCV was reported for Leaf area index, days to 50% heading, chlorophyll content, days to maturity and lowest chlorophyll fluorescence. Safi et al., (2017), Otayk Z.M., (2019), Singh, A. G., & Sharma, A. K. (2021) and Regmi et al., (2021) stated akin conclusions for days to 50% heading and days to maturity.

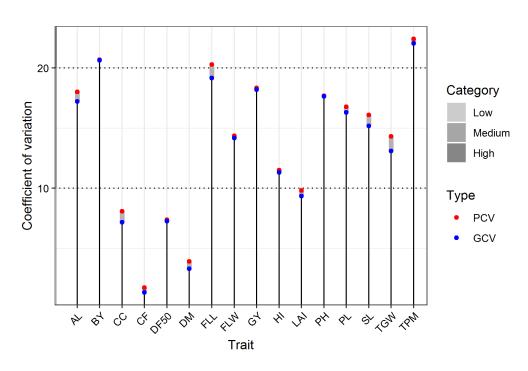


Fig. 2 Phenotypic and Genotypic Coefficient of Variability for various characters

Table 2 Details of genetic variability parameters for all the quantitative traits

S. N.	Characters	Vp	Vg	Coefficient of Variation (%)		h <sup>2</sup> (bs)	GA	(GAM)
				PCV	GCV			

1	DH	48.74	47.18	7.39	7.27	96.8	13.94	14.76
2	DM	24.53	17.62	3.91	3.31	71.83	7.34	5.79
3	CF	0.00017	0.0001	1.73	1.34	60.04	0.02	2.15
4	LAI	0.03	0.02	9.83	9.35	90.51	0.31	18.35
5	CC	8.05	6.34	8.09	7.18	78.77	4.61	13.14
6	PH	214.76	213.55	17.68	17.63	99.44	30.06	36.27
7	FL L	12.77	11.4	20.29	19.17	89.25	6.58	37.35
8	FLW	0.06	0.05	14.37	14.19	97.48	0.47	28.89
9	SL	2.78	2.47	16.1	15.18	88.92	3.06	29.53
10	AL	1.31	1.2	18.01	17.21	91.35	2.16	33.94
11	PL	27.68	26.18	16.78	16.32	94.59	10.27	32.74
12	TPM	394.66	381.52	22.41	22.04	96.67	39.62	44.7
13	TGW	34.05	28.49	14.32	13.1	83.67	10.07	24.72
14	GY	1843.43	1816.48	18.34	18.21	98.54	87.28	37.28
15	BY	14680.5	14620.1	20.68	20.63	99.59	248.93	42.48
16	HI	0.0022	0.0021	11.51	11.32	96.67	0.09	22.96

DH –Days to 50% heading, DM-Days to maturity, FW-Flag leaf width, FL-Flag leaf length, AL-Awn length, SL-Spike length, PL-Peduncle length, TPM-Effective Tillers number per meter, PH-Plant height, LAF- leaf area index, CF-chlorophyll fluorescence, CC-chlorophyll content, TGW-1000- grain weight, GY-Grain yield per row, BY- Biological yield per row, HI-Harvest index.

For all the traits studied, the magnitude of phenotypic coefficient of variation (PCV) was greater than the genotype coefficient of variation (GCV) which indicated the influence of environmental factors on their expression. For biological yield row<sup>-1</sup>, effective number of tillers metre-1 and flag leaf length, reported the maximum PCV(Fig. 2), which was consistent with the study of **Dutamo**, (2015), **Arya** *et al.*, (2017), **Alemu** *et al.*, (2019) and **Kaur** *et al.*, (2019) for biological yield row<sup>-1</sup> and effective number of tillers metre-1. Phenotypic coefficient of variation was moderate for grain yield row<sup>-1</sup>, awn length, plant height,

peduncle length, spike length, flag leaf width, 1000-grain weight and harvest index, which is in agreement with the findings of **Regmi** et al., (2021) for plant height, peduncle length and spike length, **Singh, A. G., & Sharma, A. K.** (2021) for spike length and 1000-grain weight, **Safi** et al., (2017) for peduncle length and harvest index and **Alemu** et al., (2019) for harvest index and 1000-grain weight. Phenotypic coefficient of variation was low for leaf area index, chlorophyll content, days to 50% heading and days to maturity which is in accordance with the findings of **Safi** et al., (2017), **Bayisa** et al., (2020), **Singh, A. G., & Sharma, A. K.** (2021) and **Regmi** et al., (2021), for days to 50% heading and days to maturity.

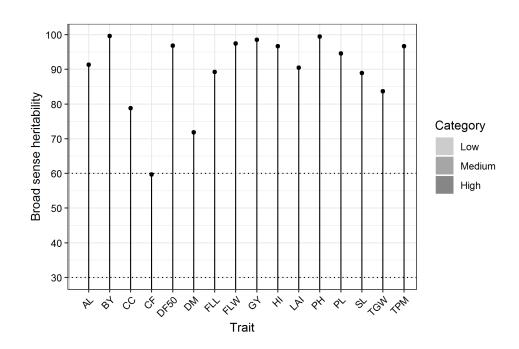


Fig. 3 Broad Sense Heritability for various characters

Estimation of heritability becomes essential as the genotypic coefficient of variation itself does not offer full scope to estimate the variation that is heritable or environmental. High heritability was observed for all the traits, except chlorophyll fluorescence representing that all of the traits have a strong genetic basis. and was less impacted by environmental factors and the likelihood of selection progress. (Fig. 3). The results obtained are consistent with those of Otayk Z.M., (2019), Singh, A. G., & Sharma, A. K. (2021) and Regmi *et al.*, (2021) for days to 50% heading, days to maturity and plant height. Dutamo, (2015) and Wahidy *et al.*, (2016) concluded similar results for harvest index, grain yield row<sup>-1</sup> and biological yield row<sup>-1</sup>. Dutamo, (2015), and Elahi *et al.*, (2020) for 1000-grain weight, Bayisa*et al.*, (2020) and Elahi *et al.*, (2020) for spike length, Singh, A. G., & Sharma, A. K. (2021) for effective number of tillers metre-1, Safi *et al.*, (2017) for peduncle length.

High value (more than 20%) of genetic advance % mean was reported for effective number of tillers metre-<sup>1</sup>, followed by biological yield row<sup>-1</sup>, flag leaf length, grain yield row<sup>-1</sup>, plant height, awn length, peduncle length, spike length, flag leaf width, 1000-grain weight and harvest index (**Fig. 4**).**Jan and Kashyap** (2018) for biological yield row<sup>-1</sup>, grain yield row<sup>-1</sup>, effective number of tillers metre-1 and 1000-grain weight, **Kumar** *et al.*, (2013), **Wahidy** *et al.*, (2016), **Arya** *et al.*, (2017) and **AL-Otyak Z.M.** *et al.*, (2019) also reported high heritability for plant height. **Rajput**, (2018) and **Alemu** *et al.*, (2019) for harvest index. Moderate values were reported (10-20%) for harvest index, leaf area index ,days to 50% heading and chlorophyll content. Similar conclusions were also conveyed by **Kumar** *et al.* (2013), **Wahidy** *et al.* (2016), **Berhanu** *et al.* (2017) for 50% days to heading, whereas, lowest values (<10%) of genetic advance as per cent of mean was showed for days to maturity, chlorophyll fluorescence. Similar results were observed by **Kumar** *et al.* (2013), **Arya** *et al.* (2017), **Sabit** *et al.* (2017), **Jan & Kashyap** (2018), **Rajput** *et al.* (2018) for days to maturity.

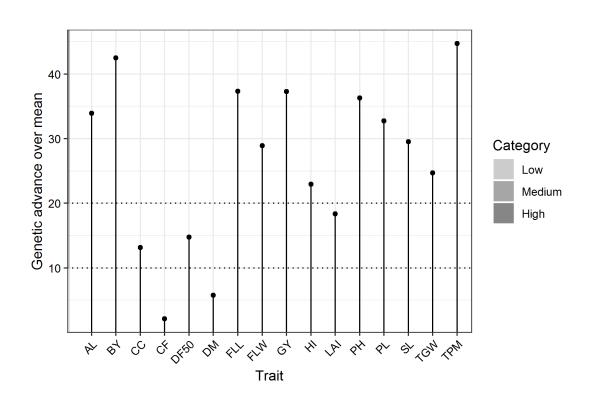


Fig. 4 Genetic advance as per cent of mean for various characters

High heritability coupled with high genetic advance per cent of mean was observed for effective tillers per meter, followed by biological yield per row, flag leaf length, grain yield per row, plant height, awn length, peduncle length, spike length, flag leaf width, 1000- grain weight, harvest index. Selective

breeding might be successful for these traits, as their heritability is likely owed to additive gene effects. In consonance with current result, **Kumar** *et al.*, (2013)reported effective number of tillers metre-1, followed by biological yield row<sup>-1</sup>, grain yield row<sup>-1</sup>, plant height, spike length, 1000-grain weight and harvest index. **Arya et al.** (2017) and **Sabit et al.** (2017) conclude similar results for 1000- grain weight, biological yield per row, flag leaf length, grain yield per row and including effective tillers per meter by **Kumar** *et al.* (2013) and **Jan & Kashyap** (2018). High heritability combined with moderate genetic advance per cent of mean was exhibited by leaf area index, days heading and chlorophyll content. High and moderate heritability along with low genetic advance per cent of mean was observed for days to maturity, chlorophyll fluorescence respectively. These findings are in accordance with those of **Kumar** *et al.* (2013), **Jan and Kashyap** (2018), and **Regmi** *et al.* (2021).

## 4.4 Correlation Analysis

Correlation studies enable the identification of interrelated characters based on those if the selection is made there are high chances for improving other related characters. Association between traits were worked out among all the characters under study and have been presented in **Table 3**.

. In consonance with the current result, **Kumar** *et al.*, **(2013)** reported effective number of tillers metre-1, followed by biological yield row<sup>-1</sup>, grain yield row<sup>-1</sup>, plant height, spike length, 1000-grain weight and harvest index. **Arya** *et al.*, **(2017)**, **Sabit** *et al.*, **(2017)**, **Jain** and **Kashyap (2018)** and **Kumar** *et al.*, **(2013)** reported similar results for 1000-grain weight, biological yield row<sup>-1</sup>, flag leaf length, grain yield row<sup>-1</sup> and effective number of tillers metre-1. High heritability combined with moderate genetic advance percent of mean was exhibited by leaf area index, days to 50% heading and chlorophyll content. High and moderate heritability along with the low genetic advance percent of mean was observed for days to maturity. These findings are in accordance with those of **Kumar** *et al.*, **(2013)**, **Jan** and **Kashyap (2018)**, and **Regmi** *et al.*, **(2021)**.

Table 3 Simple correlation analysis between sixteen characters in wheat

	DH	DM	ELI	ELW	АТ	CI	PL	TDM	DII	1 41	CE	CC	TGW	CV	DV	111
	DH	DM	FLL	FLW	AL	SL	PL	TPM	PH	LAI	CF	CC	IGW	GY	BY	HI
DH	1															
DM	0.853**	1														
FLL	-0.127	-0.083	1													
FLW	-0.246**	-0.192**	0.602**	1												
AL	0.212**	0.170*	0.281**	0.169*	1											
SL	0.190**	0.181**	0.185**	0.152*	-0.200**	1										
PL	-0.292**	-0.173*	0.446**	0.271**	0.024	0.073	1									
TPM	-0.192**	-0.189**	0.018	0.047	0.047	-0.145*	0.157*	1								
PH	-0.107	-0.019	0.127	-0.028	-0.11	0.062	0.716**	0.174**	1							
LAI	-0.242**	-0.197**	0.057	-0.011	0.015	-0.098	0.121	0.063	0.128	1						
CF	-0.136*	-0.102	0.057	$0.140^{*}$	0.04	0.023	0.109	0.017	0.058	-0.069	1					
CC	0.033	0.041	-0.028	-0.053	0.035	0.064	-0.057	-0.085	-0.077	-0.058	0.074	1				
TGW	-0.083	-0.118	-0.133*	$0.140^{*}$	-0.095	0.064	-0.055	0.093	-0.08	0.077	0.002	-0.078	1			
GY	-0.291**	-0.304**	-0.059	0.137*	-0.025	-0.078	0.111	0.328**	0.121	0.117	0.078	-0.09	0.476**	1		
BY	-0.161*	-0.160*	-0.032	0.05	-0.023	-0.08	0.121	0.341**	0.209**	0.088	0.08	-0.05	0.356**	0.805**	1	
HI	-0.165*	-0.175**	-0.019	0.115	0.003	-0.002	-0.019	-0.077	-0.144*	0.023	-0.023	-0.074	0.122	0.133*	-0.470**	1

DH –Days to 50% heading, DM-Days to maturity, FW-Flag leaf width, FL-Flag leaf length, AL-Awn length, SL-Spike length, PL-Peduncle length, TPM-Effective Tillers number per meter, PH-Plant height, LAF- leaf area index, CF-chlorophyll fluorescence, CC-chlorophyll content, TGW-1000- grain weight, GY-Grain yield per row, BY- Biological yield per row, HI-Harvest index.

<sup>\*</sup>Significant at 5 per cent level

<sup>\*\*</sup>Significant at 1 per cent level

In the present investigation, the results obtained from the correlation coefficient revealed that days to 50% heading showed significant and positive association with days tomaturity (0.853\*\*), awn length (0.212\*\*), spike length (0.190\*\*). Flag leaf width had significant and positive correlation with flag leaf length (0.602\*\*), spike length (0.152\*), awn length (0.169\*), peduncle length (0.271\*\*), 1000-grain weight (0.140\*) and grain yield row<sup>-1</sup> (0.137\*). Effective tillers meter<sup>-1</sup> exhibited significant positive correlation with plant height (0.174\*\*), peduncle length (0.157\*) grain yield row<sup>-1</sup> (0.328\*\*), biological yield row<sup>-1</sup> (0.341\*\*), whereas significant and negative association with 50% heading, (-0.192\*\*), days to maturity (-0.189\*\*), spike length (-0.145\*). 1000-grain weight exhibited significant positive and significant correlation with flag leaf width (0.140\*) and biological yield row<sup>-1</sup> (0.356\*\*).

Grain yield row<sup>-1</sup> exhibited the highest positive and significant correlation with flag leaf width (0.137\*), effective number of tillers metre-1 (0.328\*), biological yield row<sup>-1</sup> (0.805\*), harvest index (0.133\*) and 1000-grain weight (0.476\*\*) indicating that an increase in these component traits simultaneously will help to improve economic grain yield in wheat. Pooja et al., (2018), Rajput (2018), Kaur et al., (2019), Anubhav et al., (2020), Rai et al., (2020), Kumar et al., (2021) and Tsegaye et al., (2021) also identified the above characteristics as an important component of grain yield in wheat.

#### **CONCLUSION**

Plant genetic resources are characterised to aid in the identification of attribute donors for use in breeding programs. For the different characteristics under examination, the genotypes utilised in the study revealed a lot of variability and connection among themselves.

Grain yield, biological yield, effective number of tillers metre-1 and 1000-grain weight are more variable characteristics among these genotypes, according to the current study. More emphasis should be placed on those traits that have a direct or indirect impact on seed yield to enhance seed production. To do so, correlation is used to determine the link between the yield and the characteristics that contribute to the yield. Characters such as flag leaf width, effective number of tillers metre-1, biological yield row<sup>-1</sup>, harvest index and 1000-grain weight were recognized as the main selection criteria for improving seed yield in wheat, as these characters showed a strong positive correlation as well as high positive direct effects on seed yield plant<sup>-1</sup>. Therefore, when establishing a selection strategy for breeding high yielding wheat varieties, these traits or components should be given due consideration.

#### **Conflict of Interest**

The authors would hereby like to declare that there is no conflict of interest that could possibly arise.

### Acknowledgment

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