SCREENING OF WHEAT GENOTYPES FOR GENETIC DIVERSITY AND AGRO-MORPHOLOGICAL TRAITS TO ENHANCE NUTRITIONAL QUALITY

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Abstract

Wheat, a staple crop crucial for global food security, often suffers from deficiencies in essential micronutrients like zinc (Zn) and iron (Fe), leading to widespread malnutrition. Enhancing the nutritional quality of wheat through genetic diversity and agro-morphological trait selection is essential to meet the increasing demand for nutrient-rich food. To address these deficiencies, this study aimed to screen wheat genotypes for genetic diversity and agro-morphological traits, which are critical for enhancing the nutritional quality of wheat. The experiment was designed to evaluate the impact of different treatments (Normal, Soil (Fe+Zn), and Soil+Foliar (Fe+Zn)) on these traits. A total of 125 wheat genotypes were evaluated during the 2019-20 growing season at the University of Agriculture Faisalabad. The experiment employed a randomized complete block design with two replications for each treatment. Data were collected on various morphological and yield-related traits, and statistical analyses, including ANOVA and Principal Component Analysis (PCA), were conducted to identify significant differences and key contributing traits. The results revealed non-significant differences in flag leaf area among treatments but significant differences in plant height, with the Soil (Fe+Zn) treatment yielding the tallest plants. The highest plant height was observed in genotype G121, reaching 168.00 cm. In terms of spike length, significant differences were observed among treatments, with the maximum spike length recorded in genotype G13 at 13.19 cm under the Soil+Foliar (Fe+Zn) treatment. Moreover, the highest grain yield per plant was observed in genotype G56, which achieved 29.97 g under the Soil+Foliar (Fe+Zn) treatment. PCA identified spike length, spikelets per spike, 1000-grain weight, plant height, and number of grains per spike as key contributors to genotype variation, explaining 55.4% of the total variation. The study successfully identified 15 wheat genotypes with superior agro-morphological traits from this screening experiment, which have been selected for crossing in future breeding programs to enhance the nutritional quality and yield of wheat.

Keywords: Wheat Genotypes, Genetic Diversity, Agro-Morphological Traits, Nutritional Quality, Micronutrient Deficiency, Grain Yield, Breeding Programs.

1. INTRODUCTION

Wheat (*Triticum aestivum* L.) is a globally cultivated staple crop, covering 219 million hectares and producing 765.41 million metric tonnes annually (FAOSTAT, 2020). In country like Pakistan, wheat is vital to agriculture, with 9.6 million hectares yielding 31.4 million tonnes in 2023-24 (Govt. of Pakistan 2023-24). However, wheat is naturally deficient in essential micronutrients such as zinc (Zn) and iron (Fe), which are crucial for human health. This deficiency contributes to malnutrition, affecting billions globally, particularly in developing countries where wheat is a primary dietary component (WHO, 2016; Welch and Graham 2004).

To address these deficiencies, biofortification strategies, including agronomic practices and plant breeding, have been employed to enhance the nutritional quality of wheat (Cakmak 2009; Borril et al. 2014). This is particularly important as the global population continues to rise, intensifying the demand for nutrient-rich food crops (United Nations, 2012; Huang et al. 2002). Iron and zinc deficiencies are prevalent, leading to significant health issues such as anemia and impaired immune function (Stevens et al., 2013; Prasad, 2014).

Genetic diversity plays a critical role in improving wheat's adaptability and yield across various environments. Wide genetic bases enable wheat genotypes to perform better under diverse environmental conditions, and different agro-morphological traits, such as the number of fertile tillers, flag leaf size, and grain weight, significantly influence yield. Methods like principal component analysis (PCA) and cluster analysis are vital for identifying genetic diversity, selecting parent lines, and understanding evolutionary paths and environmental interactions (Tahmasebi et al., 2021).

However, the loss of genetic variability due to factors like inbreeding, genetic drift, and selection can lead to an imbalanced ecosystem and decreased crop performance (Gepts, 2006; Bapela et al., 2022). Understanding and maintaining genetic variability is crucial for the evolution and survival of wheat species, especially given the increasing global demand for wheat (Sajjad et al. 2011). Screening for genetic diversity in wheat germplasm is the first step in developing hybrid cultivars with improved traits, which can be utilized more effectively when genetic diversity patterns are well understood (Maniee et al. 2009; Masood et al. 2014; Kumar et al. 2014).

High variability in yield-attributing traits among parents and hybrids indicates the potential for direct selection of better-performing segregants. Heterosis studies further highlight that genetic diversity among parents can lead to significant improvements in plant architecture and yield, although not all traits exhibit heterosis (Amin et al. 2014; Fu et al. 2014). Therefore, strategic selection and hybridization, grounded in a thorough understanding of genetic diversity, are essential for wheat improvement programs (Jain and Sastry 2012; Abbas et al. 2008).

The primary objectives of this research were to screen wheat genotypes for genetic diversity and identify those with superior agro-morphological traits that contribute to

enhanced nutritional quality. The study aimed to select genotypes for breeding programs to develop wheat genotypes with improved nutritional profiles, while also exploring correlations between yield-related traits and genetic variability.

2. MATERIALS AND METHODS

2.1 Study Area and Experimental Design

The study was conducted during the 2019-20 growing season at the experimental fields of the Department of Plant Breeding and Genetics, University of Agriculture Faisalabad, Pakistan. The primary objective was to screen a diverse set of wheat (*Triticum aestivum* L.) genotypes for key yield-related traits. The experiment was comprised of three treatments: Normal (Control), Soil (Fe+Zn), and Soil+Foliar (Fe+Zn) with two replications to minimize environmental variability and ensure reliable results. These treatments were designed to assess the impact of varying levels of micronutrient supplementation on the wheat genotypes.

Each experimental plot measured one meter in length, with seeds sown at an inter-row spacing of 30 cm and an inter-plant spacing of 15 cm. This planting arrangement was selected to optimize growth conditions and maintain uniformity across experimental units.

2.2 Plant Material

A total of 125 wheat genotypes were used in the study. These genotypes were previously collected and characterized based on their morphological and biochemical diversity. Twelve genotypes were selected as lines (female parents) based on their high yield potential, and three genotypes were selected as testers (male parents) based on their superior performance in previous screenings. The selection was informed by data obtained during the 2018-19 growing season.

2.3 Sowing and Cultural Practices

Sowing was performed during the Rabi season of 2019-20 using a dibbler to ensure precise seed placement and consistent spacing. Two seeds were sown per hill to secure germination, and after germination, seedlings were thinned to one per hill to maintain a uniform plant population. Standard agronomic practices, including regular irrigation, fertilization, and weed control, were followed throughout the growing season to promote optimal plant growth.

2.4 Data Collection

2.4.1 Morphological and Yield-Related Traits:

2.4.1.1 Plant height (cm): Plant height was measured from the base of the plant to the tip of the spike, excluding the awns, using a meter rod. Measurements were taken from five randomly selected plants per plot, and the average height was calculated for each genotype.

2.4.1.2 Peduncle length (cm): The peduncle length, defined as the distance from the last node to the base of the spike, was measured on the main tiller of five selected plants per plot using a meter rod.

2.4.1.3 Flag leaf area (cm²): Flag leaf area was calculated by measuring the length and width of the flag leaf at three points (base, middle, and tip). The area was then estimated using the formula: Flag leaf area = Length \times Width \times 0.74. Measurements were taken from five plants per plot, and the average was calculated.

2.4.1.4 Number of tillers per plant: The total number of productive tillers per plant was counted after the completion of the vegetative phase. Data were recorded from five plants per plot, and the average number of tillers per plant was calculated.

2.4.1.5 Number of spikelets per spike: The number of spikelets on the main spike of each selected plant was counted. Data were recorded for five spikes per plot, and the average was determined.

2.4.1.6 Spike length (cm): Spike length, excluding awns, was measured from the base to the tip of the spike using a measuring tape. Measurements were taken from five plants per plot, and the average spike length was calculated.

2.4.1.7 Grains per spike: The total number of grains in the main spike was counted after threshing the spike. Data were recorded for five spikes per plot, and the average number of grains per spike was calculated.

2.4.1.8 1000-grain weight (g): The weight of 1000 grains was measured using an electronic balance. Grain samples were collected from each plot, and three replicates were weighed to calculate the average 1000-grain weight.

2.4.1.9 Grain yield per plant (g): Grain yield per plant was calculated by weighing the total grains produced by a single plant after threshing. Data were recorded from five plants per plot, and the average grain yield per plant was calculated.

2.5 Statistical Analysis

2.5.1 Analysis of Variance (ANOVA):

A one-way ANOVA was performed to assess the significance of differences among the 125 wheat genotypes for each measured trait. The significance of the results was tested at 5% (p < 0.05) and 1% (p < 0.01) levels of probability. The F-ratio was calculated to compare the variance among the genotypes to the variance within the genotypes.

2.5.2 Pearson Correlation Coefficient:

The Pearson correlation coefficient (r) was calculated to examine the relationships between the various yield-related traits. This analysis provided insights into the degree and direction of linear relationships between the traits, helping to identify traits that are positively or negatively correlated.

2.5.3 Principal Component Analysis (PCA):

PCA was conducted to reduce the dimensionality of the data and to identify the most significant traits contributing to the variation among the wheat genotypes. The data for all measured traits were standardized to have a mean of zero and a standard deviation of one. This standardization ensured that all traits contributed equally to the analysis, regardless of their original measurement scales.

The PCA was performed using the correlation matrix of the standardized data. The correlation matrix was chosen because it accounts for the relationships between traits while removing the effects of different measurement scales.

Eigenvalues and Eigenvectors: The eigenvalues and eigenvectors were computed for each principal component. Eigenvalues represent the amount of variance explained by each principal component, while eigenvectors indicate the direction of the maximum variance in the data.

Selection of Principal Components: Principal components with eigenvalues greater than 1 were considered significant and retained for further analysis. These components were assumed to capture the most substantial variability in the dataset.

Visualization: A scree plot was generated to visualize the variance explained by each principal component. This plot helped determine the number of principal components to retain by identifying the point at which the explained variance starts to diminish. A biplot was constructed to provide a graphical representation of the relationships between the genotypes and the traits. This plot displayed the first two principal components, which typically capture most of the variability in the data, allowing for a visual inspection of genotype clustering and trait contributions.

Cluster Analysis:

Hierarchical clustering was performed using Ward's method, with the Euclidean distance as the measure of similarity. A dendrogram was generated to illustrate the clustering pattern of the genotypes, helping to identify groups of genotypes with similar trait performance.

3. RESULTS

3.1 Morphological characteristics

3.1.1 Flag leaf area (cm²): The analysis of variance revealed non-significant differences among the treatments, wheat lines, and their interactions for flag leaf area (Table 1). In the Normal (Control) treatment, the highest flag leaf areas were observed in G75 (30.19 cm²), G78 (25.85 cm²), and G87 (24.46 cm²), while the lowest were recorded in G63 (15.18 cm²), G68 (15.60 cm²), and G30 (15.89 cm²). Under the Soil (Fe+Zn) treatment, G11 showed the largest flag leaf area at 24.32 cm², followed by G73 (24.28 cm²) and G124 (23.98 cm²), with the smallest areas in G87 (15.24 cm²), G31 (16.56 cm²), and G23 (16.98 cm²). For the Soil+Foliar (Fe+Zn) treatment, G11 again had the maximum flag leaf

area of 24.32 cm², followed by G73 (24.28 cm²) and G124 (23.98 cm²), while G113 (15.55 cm²), G111 (15.64 cm²), and G7 (16.30 cm²) had the lowest values (Figure 1). The selected parents' mean performance along with SE are given in (Figure 2).

3.1.2 Plant height (cm): The analysis of variance revealed highly significant differences among the treatments for plant height, indicating that the treatments had different effects on this trait. There was a non-significant difference among the wheat lines, suggesting that the genotypes performed similarly in terms of plant height (Table 1). However, a significant interaction between the wheat lines and the treatments was observed, indicating that the response of plant height varied depending on the genotype and the treatment applied.

In the Normal (Control) treatment, G121 exhibited the tallest plant height at 168.00 cm, followed by G21 (88.60 cm) and G46 (87.50 cm), while the shortest plant heights were recorded in G4 (76.49 cm), G27 (77.90 cm), and G69 (77.90 cm). Under the Soil (Fe+Zn) treatment, G10 showed the maximum plant height of 91.73 cm, with G19 (90.98 cm) and G9 (90.85 cm) following closely. The shortest heights in this treatment were observed in G87 (80.33 cm), G88 (80.72 cm), and G81 (80.81 cm).

For the Soil+Foliar (Fe+Zn) treatment, G119 reached the greatest height of 89.10 cm, followed by G120 (87.90 cm) and G124 (87.53 cm), while the minimum heights were recorded in G19 (77.30 cm), G70 (77.80 cm), and G13 (78.00 cm) (Figure 1). The selected parents' mean performance along with SE are given in (Figure 2).

3.1.3 Peduncle length (cm): The analysis of variance revealed non-significant differences among the treatments for peduncle length, indicating that the treatments had similar effects on this trait. Similarly, there were no significant differences among the wheat lines (Table 1). The interaction between wheat lines and treatments also showed non-significant differences. In the Normal (Control) treatment, G7 exhibited the longest peduncle length at 58.20 cm, followed by G22 (34.57 cm) and G3 (34.29 cm), while the shortest lengths were recorded in G85 (28.30 cm), G71 (28.55 cm), and G84 (28.60 cm).

Under the Soil (Fe+Zn) treatment, G7 again showed the maximum peduncle length at 34.23 cm, with G4 (33.81 cm) and G32 (33.72 cm) following closely. The shortest peduncle lengths in this treatment were observed in G71 (28.65 cm), G107 (29.22 cm), and G109 (29.22 cm). For the Soil+Foliar (Fe+Zn) treatment, G121 recorded the longest peduncle length at 34.23 cm, followed by G72 (33.71 cm) and G25 (33.46 cm), while G55 (25.25 cm), G49 (25.73 cm), and G50 (26.59 cm) had the shortest lengths (Figure 1). The selected parents' mean performance along with SE are given in (Figure 2).

3.1.4 Spike length (cm): The analysis of variance revealed highly significant differences among the treatments for spike length, indicating that the treatments had distinct effects on this trait. Significant differences were also observed among the wheat lines (Table 1). Additionally, the interaction between wheat lines and treatments showed highly significant differences, further highlighting the differential response of genotypes under varying treatment conditions. In the Normal (Control) treatment, G119 and G8 both recorded the

maximum spike length at 12.70 cm, followed by G124 (12.26 cm). The shortest spike lengths were observed in G33 (9.45 cm), G51 (9.50 cm), and G60 (9.50 cm). Under the Soil (Fe+Zn) treatment, G113 exhibited the longest spike length at 12.78 cm, followed by G77 (12.62 cm) and G34 (12.54 cm), while the shortest lengths were found in G70 (10.06 cm), G73 (10.43 cm), and G72 (10.51 cm). In the Soil+Foliar (Fe+Zn) treatment, G13 had the maximum spike length at 13.19 cm, followed by G18 (12.66 cm) and G10 (12.63 cm). The minimum spike lengths in this treatment were recorded in G116 (9.19 cm), G117 (9.40 cm), and G87 (9.54 cm) (Figure 1). The selected parents' mean performance along with SE are given in (Figure 2).

3.1.5 Spikelets per spike: The analysis of variance indicated that there were non-significant differences among the treatments for spikelets per spike, a highly significant variation was observed among the wheat lines. The interaction between wheat lines and treatments was non-significant, showing consistent treatment effects across different genotypes (Table 1).

In the Normal (Control) treatment, G32 exhibited the highest spikelets per spike at 18.40, followed by G21 (18.40) and G73 (18.20). The lowest spikelets per spike were recorded in G10 (14.24), G81 (15.00), and G9 (15.10). Under the Soil (Fe+Zn) treatment, G22 had the maximum spikelets per spike at 18.80, with G11 (18.60) and G29 (18.40) close behind. The lowest values in this treatment were observed in G75 (14.15), G76 (14.35), and G70 (15.00).

In the Soil+Foliar (Fe+Zn) treatment, G21 achieved the highest spikelets per spike at 18.80, followed by G100 (18.60) and G33 (18.60). The minimum spikelets per spike in this treatment were noted in G46 (14.45), G43 (15.40), and G8 (15.60) (Figure 1). The selected parents' mean performance along with SE are given in (Figure 2).

3.1.6 Number of grains per spike: The analysis of variance results indicated that there were highly significant differences among the wheat lines for the number of grains per spike. However, no significant differences were observed among the treatments. Additionally, the interaction between wheat lines and treatments was non-significant (Table 1). In the Normal (Control) treatment, G10 recorded the highest number of grains per spike at 76.19, followed by G85 (74.47) and G89 (74.05). The lowest numbers of grains per spike were observed in G109 (57.71), G106 (59.19), and G18 (59.20).

Under the Soil (Fe+Zn) treatment, G110 had the maximum number of grains per spike at 75.09, with G92 (74.80) and G35 (74.69) close behind. The lowest values in this treatment were seen in G63 (58.42), G78 (58.83), and G72 (61.20). In the Soil+Foliar (Fe+Zn) treatment, G62 exhibited the highest number of grains per spike at 77.31, followed by G61 (75.38) and G122 (74.20).

The minimum numbers of grains per spike in this treatment were recorded in G111 (58.30), G105 (60.50), and G104 (61.04) (Figure 1). The selected parents' mean performance along with SE are given in (Figure 2).

3.1.7. Grain yield per plant (g): The analysis of variance showed highly significant differences among the wheat lines for grain yield per plant, indicating variability in performance. The interaction between wheat lines and treatments was non-significant (Table 1). In the Normal (Control) treatment, G100 recorded the highest grain yield per plant at 21.63 g, followed by G68 (20.62 g) and G41 (20.53 g). The lowest yields were observed in G12 (12.60 g), G57 (12.71 g), and G110 (12.76 g).

Under the Soil (Fe+Zn) treatment, G108 achieved the highest yield at 22.65 g, with G81 (21.76 g) and G44 (21.06 g) close behind. The lowest yields in this treatment were found in G7 (11.58 g), G107 (12.31 g), and G3 (13.31 g). In the Soil+Foliar (Fe+Zn) treatment, G56 had the highest yield at 29.97 g, followed by G65 (27.84 g) and G57 (27.76 g). The minimum yields were recorded in G109 (13.98 g), G17 (14.16 g), and G51 (15.39 g) (Figure 1). The mean performance of the selected parents, along with the standard error (SE), is presented in the accompanying (Figure 2).

3.1.8 1000-grain weight (g): The analysis of variance indicated a highly significant difference among the treatments for 1000-grain weight. The wheat lines themselves showed no significant differences, however a significant interaction was observed between wheat lines and treatments (Table 1). In the Normal (Control) treatment, G100 recorded the highest 1000-grain weight at 43.92 g, followed by G21 (41.45 g) and G71 (41.24 g). The lowest weights were observed in G24 (31.47 g), G6 (31.63 g), and G9 (31.84 g).

Under the Soil (Fe+Zn) treatment, G108 had the maximum 1000-grain weight at 41.74 g, with G48 (41.54 g) and G64 (41.15 g) close behind. The lowest weights in this treatment were recorded in G72 (31.49 g), G105 (31.86 g), and G88 (32.32 g). In the Soil+Foliar (Fe+Zn) treatment, G5 achieved the highest 1000-grain weight at 43.21 g, followed by G10 (42.45 g) and G41 (41.42 g). The minimum weights in this treatment were observed in G60 (30.95 g), G109 (31.07 g), and G27 (31.49 g) (Figure 1). The selected parents' mean performance along with SE are given in (Figure 2).

Source	DF	FLA	PH	PL	SL	SPS	NGS	GYP	1000-GW
Rep.	1	1642.59	126.98	0.0866	63.5399	82.8075	553.256	25.278	210.548
Treatment	2	2.09ns	1144.04**	6.0029 ^{ns}	10.0432**	1.8731 ^{ns}	357.787 ^{ns}	407.888 ^{ns}	56.609 ^{ns}
Genotypes	124	8.63	58.83 ^{ns}	9.8243 ^{ns}	0.7474*	1.0791**	18.062 ^{ns}	10.37 ^{ns}	4.737 ^{ns}
Treat*Gen.	248	6.96	75.01*	10.1149 ^{ns}	0.9239**	0.6987 ^{ns}	18.554 ^{ns}	8.1 ^{ns}	4.974*
Error	374	7.6	61.64	9.6726	0.5891	0.7295	18.325	8.918	4.08

Table 1: Analysis of variance for all traits

FLA= Flag leaf area, PH= Plant height, PL= Peduncle length, SL= Spike length, SPS= Spikelet per spike, NGS= Number of grains per spike, GY= Grain yield per plant and 1000-GW= Thousand grain weight



Figure 1: Mean graph of morphological characteristics for 125 genotypes across different treatments



Figure 2: Mean graph of morphological characteristics of selected parents for different treatments

3.2. Pearson correlation

The correlation analysis revealed various relationships between the measured traits. Flag leaf area showed non-significant correlations with all other traits similarly, plant height (PH) was non-significantly associated with these traits, suggesting a lack of strong linear

relationships (Figure 3). Peduncle length exhibited a significant positive correlation with spike length ($r = 0.187^*$), indicating that longer peduncles tend to be associated with longer spikes. However, peduncle length had non-significant correlations with the other traits. Spike length also showed a significant positive correlation with peduncle length (r = 0.187^{*}) and a significant negative correlation with grain yield per plant ($r = -0.193^*$), suggesting that longer spikes might slightly reduce grain yield. Spike length had nonsignificant correlations with the remaining traits. Spikelets per spike (SPS) were nonsignificantly correlated with all other traits, indicating its relative independence. The number of tillers per plant showed a significant positive correlation with 1000-grain weight $(r = 0.221^*)$, suggesting that genotypes with more tillers might produce heavier grains, but had non-significant associations with other traits. The number of grains per spike had a significant positive correlation with 1000-grain weight (r = 0.266**) but was nonsignificantly correlated with the other traits. Grain yield per plant was significantly positively correlated with 1000-grain weight ($r = 0.182^*$) but was non-significantly associated with most other traits except for a significant negative correlation with spike length ($r = -0.193^*$). Finally, 1000-grain weight showed significant positive correlations with the number of tillers per plant ($r = 0.221^*$) and the number of grains per spike (r =0.266**), highlighting positive association with these yield-related traits, while its correlations with other traits were non-significant.

	Heatmap of Pearson Correlation and Sig. (2-tailed)											
FLA	1.00 (0.000)	-0.15 (0.090)	-0.02 (0.796)	-0.04 (0.683)	0.01 (0.918)	0.04 (0.653)	0.00 (0.974)	0.13 (0.147)	0.02 (0.854)			1.0
H	-0.15 (0.090)	1.00 (0.000)	0.07 (0.465)	-0.01 (0.904)	-0.16 (0.071)	0.11 (0.240)	-0.02 (0.860)	-0.14 (0.120)	0.04 (0.700)		-	0.8
٦. ۲	-0.02 (0.796)	0.07 (0.465)	1.00 (0.000)	0.19 (0.037)	0.05 (0.560)	-0.10 (0.244)	-0.09 (0.309)	-0.06 (0.476)	-0.15 (0.091)			0.6
SL	-0.04 (0.683)	-0.01 (0.904)	0.19 (0.037)	1.00 (0.000)	0.11 (0.217)	0.01 (0.899)	-0.07 (0.425)	-0.19 (0.031)	0.01 (0.871)		-	0.6
SPS	0.01 (0.918)	-0.16 (0.071)	0.05 (0.560)	0.11 (0.217)	1.00 (0.000)	-0.01 (0.931)	-0.13 (0.149)	-0.07 (0.435)	0.09 (0.340)		-	0.4
NTP	0.04 (0.653)	0.11 (0.240)	-0.10 (0.244)	0.01 (0.899)	-0.01 (0.931)	1.00 (0.000)	0.07 (0.435)	-0.01 (0.922)	0.22 (0.013)			
NGS	0.00 (0.974)	-0.02 (0.860)	-0.09 (0.309)	-0.07 (0.425)	-0.13 (0.149)	0.07 (0.435)	1.00 (0.000)	0.12 (0.190)	0.27 (0.003)		-	0.2
GYP	0.13 (0.147)	-0.14 (0.120)	-0.06 (0.476)	-0.19 (0.031)	-0.07 (0.435)	-0.01 (0.922)	0.12 (0.190)	1.00 (0.000)	0.18 (0.042)		-	0.0
000-GW	0.02 (0.854)	0.04 (0.700)	-0.15 (0.091)	0.01 (0.871)	0.09 (0.340)	0.22 (0.013)	0.27 (0.003)	0.18 (0.042)	1.00 (0.000)			
Ē,	FLA	PH	PL	SL	SPS	NTP	NGS	GYP	1000-GW	/		

FLA= Flag leaf area, PH= Plant height, PL= Peduncle length, SL= Spike length, NT= Number of tillers, SPS= Spikelet per spike, NGS= Number of grains per spike, GY= Grain yield per plant and TGW= Thousand grain weight

Figure 3: Pearson correlation analysis for morphological characteristics

3.3 Principle component analysis:

The Principal Component Analysis (PCA) results reveal that the first four principal components account (PC1, PC2, PC3 and PC4) showed more than 1 eigenvalue while the remaining principal components showed very low eigen-value for all the other traits (Table 2). Specifically, PC1 has an eigenvalue of 1.4113, explaining 15.7% of the total variation, making it the most influential component in distinguishing the underlying structure of the data. PC2 follows with an eigenvalue of 1.3098, contributing 14.6% to the total variation. Together, PC1 and PC2 account for 30.2% of the variation, indicating that these two components encapsulate a substantial amount of the dataset's diversity. PC3 and PC4 have eigenvalues of 1.1651 and 1.1011, respectively, contributing 12.9% and 12.2% of the total variation. The cumulative contribution of these four components amounts to 55.4%, suggesting that they capture the most critical variation patterns among the traits.

The remaining components, PC5 through PC9, contribute progressively smaller amounts to the total variation, with eigenvalues ranging from 0.9635 to 0.6818. These components explain between 7.6% and 10.7% of the variation individually, with the cumulative explained variation reaching 100% by PC9. This indicates that while the first four components encapsulate the major patterns within the data, the subsequent components still account for important but smaller variations, ensuring that the full breadth of the data's diversity is captured across all nine components.

The PCA revealed that spike length (SL), spikelets per spike (SPS), and 1000-grain weight (TGW) were key in PC1 (15.7% variation). PC2 (14.6%) was driven by plant height (PH) and number of grains per spike (NG). PC3 (12.9%) and PC4 (12.2%) were influenced by peduncle length (PL), number of tillers (NT), and flag leaf area (FLA). The remaining components had smaller contributions, with FLA, NG, PH, and SL being notable across them, highlighting these traits as central to the variation among genotypes.

Variable	Eigenvalue	% Total Variation	Cumulative eigenvalue	% Cumulative	
PC1	1.4113	15.7	0.157	15.7	
PC2	1.3098	14.6	0.302	30.2	
PC3	1.1651	12.9	0.432	43.2	
PC4	1.1011	12.2	0.554	55.4	
PC5	0.9635	10.7	0.661	66.1	
PC6	0.8637	9.6	0.757	75.7	
PC7	0.8006	8.9	0.846	84.6	
PC8	0.7032	7.8	0.924	92.4	
PC9	0.6818	7.6	1	100	

Table 2: Eigen analysis of the correlation matrix

Variable	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9
FLA	0.234	-0.163	0.016	0.49	0.666	-0.439	0.07	0.008	-0.191
PH	-0.048	0.656	0.14	-0.011	0.074	-0.033	-0.541	-0.02	-0.497
PL	-0.114	0.183	0.647	-0.316	0.105	-0.189	0.514	-0.344	-0.077
SL	0.536	0.09	0.388	-0.171	-0.098	-0.181	-0.085	0.645	0.244
SPS	0.502	-0.28	-0.041	0.009	-0.523	-0.176	0.048	-0.27	-0.537
NT	-0.151	0.326	-0.542	-0.236	-0.094	-0.504	0.398	0.29	-0.127
NG	0.399	0.348	-0.156	0.138	0.166	0.631	0.47	0.075	-0.158
GY	-0.075	-0.421	-0.044	-0.599	0.383	0.211	-0.071	0.262	-0.438
TGW	0.45	0.137	-0.298	-0.444	0.271	-0.104	-0.211	-0.484	0.36

Table 3: Eigenvectors of principal component analysis

FLA= Flag leaf area, PH= Plant height, PL= Peduncle length, SL= Spike length, NT= Number of tillers, SPS= Spikelet per spike, NGS= Number of grains per spike, GY= Grain yield per plant and TGW= 1000-grain weight

Scree Plot

The Scree plot (Figure 4) revealed the contribution of variability related with each principal component. The Scree plot resulted in a graph between the number of PCs and eigenvalues. The PC 1 showed 15.7 % variability with 1.411 eigenvalues. The scree plot indicated that all the PCs are showing a pattern of variance through a curve. The PC2, PC3, and PC4 showed a considerable variance of 14.6%, 12.9 % and 12.2% with an eigenvalue of 1.309, 1.165 and 1.101 respectively. The PC5, PC6, PC7 and PC8 depict very low contributions to variability and eigenvalue below than 1. The PC9 showed a very low contribution with a minimum eigenvalue of 0.681.



Figure 4: Scree plot of Principal component analysis for all traits



Figure 5: Biplot of principal component analysis for all traits

4. DISCUSSION

The current study investigated the effects of various treatments on morphological characteristics and yield components of wheat genotypes, focusing on traits such as flag leaf area, plant height, peduncle length, spike length, spikelets per spike, number of grains per spike, grain yield per plant, and 1000-grain weight. The non-significant differences in flag leaf area among treatments, wheat genotypes, and their interactions suggest that this trait is relatively stable across different environmental conditions and genetic backgrounds. In contrast, plant height showed highly significant differences among treatments and a significant interaction between wheat lines and treatments, indicating that Fe+Zn application, particularly in soil, may enhance elongation growth, as suggested by Cakmak (2008) and confirmed by Bouis and Welch (2010), who demonstrated the role of micronutrients in promoting plant growth. Peduncle length, however, was not significantly influenced by the treatments, which aligns with previous studies highlighting its stability across environments. Spike length exhibited highly significant differences among treatments and wheat genotypes, with a notable negative correlation with grain yield per plant, suggesting a trade-off that may be influenced by resource allocation and competition for assimilates within the plant, as reported by Fischer (2011). Spikelets per spike, while showing non-significant differences across treatments, varied significantly among wheat lines, underscoring the genetic control over this trait, as has been demonstrated in studies focusing on the genetic determinants of spikelet number in wheat (Hnizil et al., 2024). The number of grains per spike showed strong genetic control with significant positive correlations with 1000-grain weight, indicating its importance in yield determination results align with Ullah et al. (2018). Grain

yield per plant was significantly influenced by 1000-grain weight, reinforcing the importance of grain size in overall yield (Wang et al., 2015). The significant differences in 1000-grain weight among treatments and its correlation with other yield components further underscore its critical role in wheat productivity (Rehan et al., 2024). Principal component analysis revealed that traits like spike length, spikelets per spike, 1000-grain weight, plant height, and number of grains per spike were key contributors to the variation among wheat lines, indicating their importance in distinguishing genotypic responses to the treatments. This finding aligns (Davesh et al., 2019) emphasizing the role of these traits in the overall yield architecture of wheat.

5. CONCLUSION

This study effectively screened 125 wheat genotypes to identify those with superior genetic diversity and agro-morphological traits that contribute to enhanced nutritional quality. Significant variations were observed in plant height, spike length, and grain yield across different treatments, with the Soil+Foliar (Fe+Zn) treatment showing the most pronounced effects. Principal Component Analysis (PCA) identified key traits contributing to the observed genetic diversity. Based on these findings, 15 high-performing genotypes have been selected for future breeding programs aimed at improving wheat's nutritional profile, thereby addressing micronutrient deficiencies and contributing to global food security.

Conflict of Interest

The Authors have no Conflict of Interest

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