RESEARCH ARTICLE

Multivariate analysis of selected agro-morphological traits of common wheat genotypes for estimation of genetic diversity

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ABSTRACT

Estimation of genetic diversity of plant species using agro morphological parameters is of very crucial importance in plant improvement programs. In view of that common wheat (*Triticum aestivum* L.) genotypes consisting of stripe rust resistant genes were analyzed by cluster analysis, principle component analysis and coefficient of correlation analysis for determination of genetic diversity. Experiment was conducted in fields of Hazara University Mansehra Pakistan. Eight groups were categorized by cluster analysis using UPGMA clustering method. AOC-YR/ATTILA (CGSS97Y0061S-23Y-1B-1B) (Sdf31) and 10790 (sdf2) are reported as more diversified by cluster analysis. Based on principal component analysis the first five clusters showed 88.94% of genetic variation. Grains per spike contributed more (0.7) to total variation in PCI. While Harvest Index was the major contributor (0.8) to total variation in PCII. 1056 (Sdf5), AOC-YR/ATTILA (CGSS97Y0061S-32Y-1B-1B) (sdf30), 1076 (sdf9), and LALBMONO4*4/PRL//LALB (CGSS99Y00093S-2F1-22Y-9GHB-1GHB) (sdf32) were observed superior in flag leaf area, spike length, grains per spike, plant height, biological yield, plant yield and harvest index. AOC-YR/ATTILA (CGSS97Y0061S-32Y-1B-1B) (Sdf30) was found as early maturing genotype. Correlation analysis showed positive and significant association of plant yield with flag leaf area, spike length and biological yield, while it is highly negatively correlated to no. of days to maturity. More diversified genotypes identified in this studies can be used in future breeding programs along with utilization of superior genotypes in hybridization.

Keywords: Cluster analysis; Correlation; Genetic diversity; Principal component analysis; Wheat.

INTRODUCTION

Globally, wheat production is of high demand to be used as primary food and livestock feed. Wheat demand is increasing day by day with increasing population of the world, instead, its production and quality is continuously rendered directly or indirectly by many biotic and abiotic factors. Plant scientists and wheat breeders are focusing to overcome the stressors by performing and launching different breeding experiments and programs for wheat improvement (Martin et al., 2008; Van de Wouw et al., 2010). The breeding programs are initiated with quantification of genetic variation of agro morphological traits of a diverse population and evaluation and utilization of those genotypes further for crop improvements (Nawaz et al., 2013). Genetically more diverse genotypes always benefits for transgressive segregation and higher heterosis in case

of hybridization (Joshi et al., 2004; Joshi & Dhawan, 1966; Anand & Murrty, 1968). Estimation of genetic distance through different agronomic traits or by application of different molecular markers RFLP, SSR, AFLP and RAPD is of utmost importance (Anas & Yoshida, 2004; Helguera et al., 2005; Gashaw et al., 2007; Li et al., 2009; Zeb et al., 2009; Ahmed et al., 2010; Colomba & Gregorini, 2011; Al-Fares & Abu-Qaoud, 2012).

Presently, a number of appropriate approaches like principal component analysis (PCA), factor analysis, cluster analysis and correlation coefficient analysis are trending for hybridization and estimation of genetic diversity (Mostafa et al., 2011). Previous work of many authors can be practically applied to evaluate genetic diversity among wheat genotypes and find their relationships on basis of morphological traits by using cluster and principal

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Table 1: Sixty three wheat genotypes (research code "sdf" is representing genotypes in study)

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|----|--|-------|---|--|--|--|--|--|--|
| | Genotypes | s.no. | Genotypes | | | | | | |
| 1 | 1216 (sdf1) | 33 | AOC-YR*3//LALBMONO1*4/PVN (CGSS01Y00011T-099B-27Y) (sdf33) | | | | | | |
| 2 | 10790 ((sdf2) | 34 | AOC-YR*3//LALBMONO1*4/PVN (CGSS01Y00011T-099B-16Y) (sdf34) | | | | | | |
| 3 | Raskoh (sdf3) | 35 | AOC-YR*3/PASTOR (sdf35) | | | | | | |
| 4 | 1050(sdf4) | 36 | AOC-YR*3//LALBMONO1*4/PVN (CGSS01Y00011T-099B-37Y) (sdf36) | | | | | | |
| 5 | 1056(sdf5) | 37 | AOC-YR*3/3/ALTAR84/AE.SQ//OPATA (CGSS00Y00204T-099M-33Y) (sdf37) | | | | | | |
| 6 | 1072(sdf6) | 38 | AOC-YR*3/3/ALTAR84/AE.SQ//OPATA (CGSS00Y00204T-099M-32Y) (sdf38) | | | | | | |
| 7 | 1099(sdf7) | 39 | AOC-YR*3/3/ALTAR84/AE.SQ//OPATA (CGSS00Y00204T-099M-20Y) (sdf39) | | | | | | |
| 8 | 1055(sdf8) | 40 | AOC-YR*3/3/ALTAR84/AE.SQ//OPATA (CGSS00Y00204T-099M-3Y) (sdf40) | | | | | | |
| 9 | 1076(sdf9) | 41 | YRCV/6*AOC (sdf41) | | | | | | |
| 10 | 1018(sdf10) | 42 | POLLMER_2.1.1 (CTY88.547-22RES-1M-0Y-2M-1Y-0M-1B-0Y) (sdf42) | | | | | | |
| 11 | Fs x barani83(sdf11) | 43 | SUPER KAUZ (CM67458-4Y-1M-3Y-1M-3Y-0B) (sdf43) | | | | | | |
| 12 | 1294(sdf12) | 44 | OPATA M 85 (CM40038-6M-4Y-2M-1Y-2M-1Y-0B-0MEX) (sdf44) | | | | | | |
| 13 | lqbal 2002(sdf13) | 45 | SERI M 82 (CM33027-F-15M-500-0M-87B-0Y-0MEX) (sdf45) | | | | | | |
| 14 | 1121(sdf14) | 46 | PAVON F 76 (CM8399-D-4M-3Y-1M-1Y-1M-0Y-0MEX) (sdf46) | | | | | | |
| 15 | Sutlaj 86(sdf15) | 47 | YRSP/6*AOC (CX94.14.1.15) (sdf47) | | | | | | |
| 16 | 1089(sdf16) | 48 | YR27/6*AOC (-386) (sdf48) | | | | | | |
| 17 | 1051(sdf17) | 49 | YR26/3*AOC (CX96.17.1) (sdf49) | | | | | | |
| 18 | 1090(sdf18) | 50 | YR24/3*AOC (CX96.1.3.12) (sdf50) | | | | | | |
| 19 | 1019(sdf19) | 51 | YR18/3*AOC (CX94.10.1.7) (sdf51) | | | | | | |
| 20 | 1098(sdf20) | 52 | YR15/6*AOC (CX89.1.1.27) (sdf52) | | | | | | |
| 21 | 1073(sdf21) | 53 | YR10/6*AOC (CX93.53.3.1) (sdf53) | | | | | | |
| 22 | 1078(sdf22) | 54 | YR9/6*AOC (CX93.24.1.22) (sdf54) | | | | | | |
| 23 | 1077(sdf23) | 55 | YR7/6*AOC (CX93.21.3.1) (sdf55) | | | | | | |
| 24 | 1081(sdf24) | 56 | YR6/6*AOC (CX94.2.2.25) (sdf56) | | | | | | |
| 25 | 1188(sdf25) | 57 | YR5/6*AOC (CX86.6.1.20) (sdf57) | | | | | | |
| 26 | AOC-YR/ATTILA (CGSS97Y0061S-148Y-1B-1B)(sdf26) | 58 | TATARA (CM85836-50Y-0M-0Y-2M-0Y-0PAK) (sdf58) | | | | | | |
| 27 | AOC-YR/ATTILA (CGSS97Y0061S-142Y-1B-1B) (sdf27) | 59 | SIETE CERROS T66 (118156-1M-2R-4M-0Y) (sdf59) | | | | | | |
| 28 | AOC-YR/ATTILA (CGSS97Y0061S-93Y-1B-1B) (sdf28) | 60 | YR1/6*AOC (CX93.51.3.3) (sdf60) | | | | | | |
| 29 | AOC-YR/ATTILA (CGSS97Y0061S- 92Y-1B-1B) (sdf29) | 61 | AVOCET+YRA (YRA) (sdf61) | | | | | | |
| 30 | AOC-YR/ATTILA (CGSS97Y0061S-32Y-1B-1B) (sdf30) | 62 | AVOCET-YRA (YRA) (sdf62) | | | | | | |
| 31 | AOC-YR/ATTILA (CGSS97Y0061S-23Y-1B-1B) (sdf31) | 63 | MOROCCO (sdf63) | | | | | | |
| 32 | LALBMONO4*4/PRL//LALB (CGSS99Y00093S-2F1-22Y -9GHB-1GHB) (sdf32) | | | | | | | | |

component analysis (Davesh et al., 2019; Beheshtizada et al., 2013). The cluster analysis (CA) is advantageous in terms that genotypes to be analyzed are grouped on the basis of complex traits instead of one character (Brown-Guedira et al., 2000). By applying principal component analysis (PCA), a number of possibly correlated variables could be transformed into a smaller number of variables called principal components (Mujaju & Chakuya, 2008). The correlation coefficient analysis demonstrates relationship between traits and provides the degree of association between various traits of a crop with crop productivity.

In the current study genetic diversity among 63 wheat (*Triticum aestivum*) genotypes was evaluated by application of multivariate analysis on important agro-morphological

traits of wheat to identify more diverse genotypes to be crossed or hybridized in future breeding programs.

MATERIAL AND METHODS

Plant material consisted sixty three wheat genotypes (possessing different stripe rust genes) including introduction from CIMMYT and local varieties established decades ago (Table 1). Field sowing was done using Randomized Complete Block Design with 30cm gap between rows, while length of row was kept about 3 meters (Fig. 1). Normal agricultural practices (irrigation and application of weedicides and fertilizers) were followed throughout the growing season till harvesting. The morphological data was collected on specific

Table 2: Clusters detail of the studied genotypes

| Clusters | Frequency | Genotype's code |
|--------------|-----------|---|
| Cluster I | 1 | sdf31 |
| Cluster II | 1 | sdf5 |
| Cluster III | 1 | sdf32 |
| Cluster IV | 34 | sdf4, sdf7, sdf9, sdf12, sdf13, sdf14, sdf15, sdf18, sdf23, sdf24, sdf25, sdf26, sdf27, sdf28, sdf29, sdf36, sdf37, sdf38, sdf41, sdf42, sdf44, sdf45, sdf46, sdf47, sdf48, sdf49, sdf50, sdf53, sdf56, sdf57, sdf60, sdf55, sdf52, sdf19, sdf20 |
| Cluster V | 4 | sdf61, sdf62, sdf63, |
| Cluster VI | 2 | sdf40, sdf43 |
| Cluster VII | 18 | sdf1, sdf3, sdf6, sdf8, sdf10, sdf11, sdf16, sdf17, sdf21, sdf22, sdf33, sdf34, sdf35, sdf39, sdf51, sdf54, sdf59, sdf58, |
| Cluster VIII | 2 | sdf2, sdf30 |

Table 3: Superior genotypes identified on basis of important agro-morphological traits

| Traits of interest | Range | Varieties identified | | |
|-----------------------|-------|----------------------------------|--|--|
| Spikes per plant | ≥7 | sdf27, sdf29, sdf40, sdf42 | | |
| Flag leaf area | ≥41 | sdf5, sdf6, sdf19, sdf50, | | |
| Spike length | ≥15 | sdf10, sdf30 | | |
| Grains per spike ≥ 81 | | sdf3, sdf17, sdf30, sdf32, sdf33 | | |
| Spikelets per spike | ≥ 25 | sdf25, sdf27 | | |
| Days to maturity | ≤169 | sdf12, sdf13, sdf30 | | |
| Plant height | ≥105 | sdf2, sdf6, sdf21, sdf30, sdf34 | | |
| Biological yield | ≥50 | sdf5, sdf6, sdf30, sdf54 | | |
| Plant yield | ≥21 | sdf5, sdf9, sdf30, sdf48, sdf54, | | |
| Thousand grain weight | ≥60 | sdf3, sdf22, sdf29 | | |
| Harvest index | ≥64 | sdf9, sdf13, sdf18, sdf31, sdf32 | | |

growth stages of wheat plant according to wheat descriptor (IBPGR, 1978). The data collected on agronomic traits from the wheat genotypes were subjected to statistical analysis using computer program PAST (Paleontological Statistics, hammer 2016) version 3.11 (Hammer, 2016).

RESULTS AND DISCUSSIONS

Cluster analysis

Cluster analysis is a modified method to classify a range of diverse varieties in different clusters on basis of their genetic relationships (Vural & Karasu, 2007). Un-weighted Pair Group Method using Arithmetic averages (UPGMA) clustering method is the most frequently used method to describe relationship among diverse genotypes.

Euclidian similarity coefficient analysis was computed for thirteen quantitative traits of wheat. It resulted in formation of 8 clusters from 63 wheat genotypes by incision on 31 unit distance on dendogram (Fig. 2). First 3 clusters consist of single genotypes. Cluster IV comprised of 34 genotypes, 4 genotypes were grouped in Cluster V. Cluster VI contain 2 genotypes. 18 genotypes were grouped in Cluster VII,



Fig 1. Experimental field of wheat (RCBD)

while only 2 genotypes were involved in formation of Cluster VIII (Fig. 2 and Table 2).

Genotypes of cluster V took maximum days to 50% heading (109), minimum number of days were counted for cluster I (101) (Fig. 3A). Spikes per plant values did not varied significantly between all clusters, only group VI genotypes revealed highest number of spikes per plant (mean value 6.5). Cluster II genotypes indicated highest values for flag leaf area (37) (Fig. 3A). Genotypes of Cluster VIII had shown superiority in Spike length (14) (Fig. 3A). Highest number of Grains per spikes was observed in cluster III (82) (Fig. 3A). Mean value for days to maturity was ranged 170-174 for all clusters except members of cluster VIII which took 178 days to be matured (Fig. 3B). Plant height was recorded as high as 110 in Cluster VIII followed by cluster VII. Cluster VIII had shown superiority in spikelet per spike (24) (Fig. 3B). Mean length of peduncle varied significantly among eight clusters (28-47.5) (Fig. 3B). Genotypes comprising cluster VI revealed maximum weight of 1000 grains which was 54gm, cluster VII had output of 49gm of 1000 grains weight while lowest figure was observed from cluster I (33). Highest values of biological yield and plant yield were indicated by Cluster II genotypes. Genotypes grouped in cluster I revealed highest harvest index value (74) (Fig. 3C).

All genotypes (Cluster II) that mature early and expressing superior yield traits in field (biological yield and plant yield) could be utilized and promoted for breeding programs for further crop improvement plans. Better genotypes regarding quality and agronomic parameters can be utilized for genes transfer techniques for crop quality improvement. No single genotype with all morphologically ideal values was identified, but gene pyramiding can assemble all desired characters from diverse genotypes to a single genotype by utilizing genotypes with important agro morphological traits identified in current research work (Table 3). Cluster analysis carried out in present studies showed significant diversity among genotypes as no duplication of data was recorded (Fig. 2) Numerous wheat scientist calculated agro-morphological diversity of wheat genotypes through Cluster analysis (Verma et al., 2014; Kumar et al., 2016).

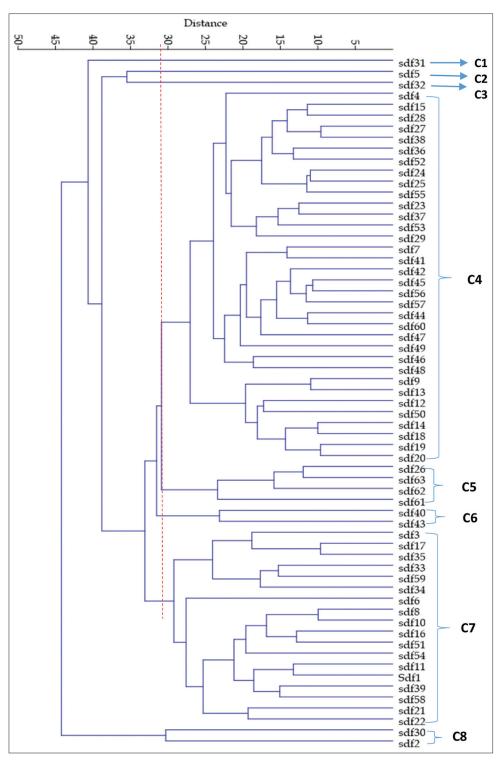


Fig 2. Dendrogram showing the relationship among 63 wheat genotypes based on 13 quantitative characters.

Principal component analysis

In current studies, PCA analysis revealed that depending on 13 vital morphological characters of wheat, first five PCs are contributing for 88.94% of total variation. PC1 was associated with 38.57% of entire variance. Highly contributing character of this component was grains

per spike (0.738) (Table 4). PCII which showed 20.128% contribution in total variance had harvest index (0.847) as major contributor to total variance. In PCIII grains per spike (0.524) was having high variance values, PCIII was contributing 13.83% to total variance. These first three PCs were associated with 72.53% of total variance. Plant height

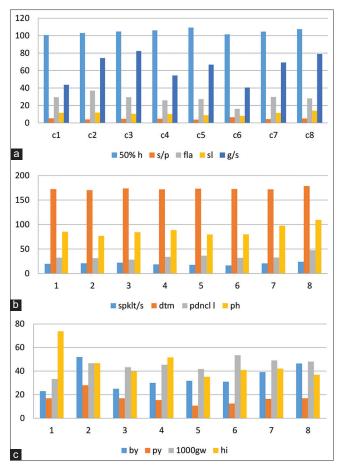


Fig 3. (A-C) Characteristic means of eight similarity clusters generated by UPGMA based on the classification of 63 genotypes of wheat (T. aestivum.L) using thirteen agro-morphological characteristics.

(0.686) was major contributing characters of PCIV which represents 10.156% variance to total variation while PCV showed 6.25% variation had 1000 grains weight (0.712) as major contributing factor. Present work is in resemblance with publication of Beheshtizadeh et al., (2013) in 2013 in which by performing PCA he found that PC I to IV were accounted for 76% of total variation and main characters associated to variation of wheat genotypes were plant yield, number of tillers, 1000 grain weight, number of grains per spike, and height of a plant. Adilova et al. (2020); also performed PCA on 25 wheat genotypes to analyze their yield data.

In current studies clustering pattern of 63 wheat genotypes by PCA elaborated genetic diversity among genotypes based on 13 morphological characters. Scator plot and biplot were graphed for first three PCs against each other to find out associations among currently studied wheat genotypes (Figs. 4 and 5). Among wheat genotypes that showed high genetic diversity in the PCI and PCII first consisted of sdf31, sdf32, sdf13, sdf9, sdf3, sdf6, sdf30 and sdf40. On the other hand, most diverse wheat varieties in the PCI and PCIII components were

sdf6, sdf22, sdf30, sdf33, sdf62, sdf19, sdf34 and sdf26 (Fig. 5).

Correlation

Correlation analysis presents the intensity of dependence (correlation) between traits. Many wheat breeders focus to enlighten the relationship of grain yield and agromorphological traits by applying simple correlation coefficients. Correlation analysis explains various characters and their association with yield. Yield is ultimate interest of crop breeders. Consequently every trait or character chosen for selection criterion must be correlated to yield. It was concluded from current studies that quantitative traits were correlated to each other either highly significantly (p<0.01), significantly (p<0.05) or non-significantly (p>0.05). Direction of correlation coefficient (positive or negative) was important to conclude the results.

In present studies sixty three wheat genotypes were analyzed for traits correlations. Values of correlation coefficient (r values) and the significance of r values are given in Table 5. According to correlation coefficient analysis spike length was positively significantly linked with flag leaf area while negatively highly significant with 50% heading and spikes per plant.

Results described in Table 5 are evident that strong significant positive relation exists among grains per spike, flag leaf area and spike length. On the other hand grains per spike is negatively significant to total spikes of a plant. There was significant positive correlation of spikelets per spike with flag leaf area, number of grains/spike and length of spike. Correlation studies of days to maturity indicated its positive significance towards days to 50% heading. Peduncle length was significantly positively associated with days to maturity.

Current studies revealed highly significantly positive association of plant height with flag leaf area, spike length, number of spikelets and grains per spike. This is in accordance with work of (Mohsin et al., 2009). The correlation observed positively significant among biological yield, length of spike, number of spikelets and grain per spike and plant height. It was also observed in present studies that plant yield was negatively linked with days to 50% heading and days to maturity, these results are in resemblance with Gashaw et al. (2007); who worked on yield related characters of wheat varieties and concluded that days to 50% heading were negatively correlated with traits linked with yield. In present studies the same trait was analyzed significantly positively correlated with flag leaf area, spike length and biological yield. Positive and significant association of 1000 grains weight was also

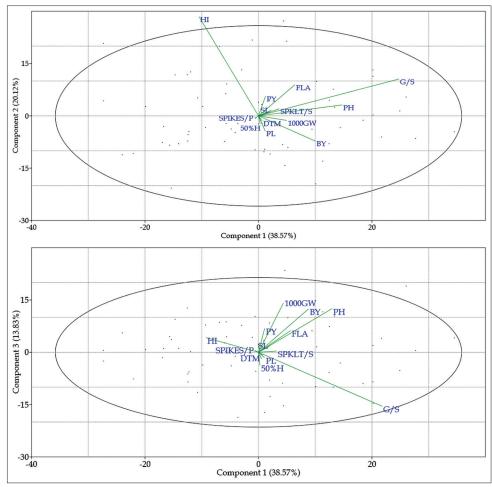


Fig 4. PCA biplots showing the involvement of 13 traits in first three PCs to total variation among 63 wheat genotypes.

Table 4: Depending on 13 morphological traits resulted Eigenvectors, eigenvalues, total variance and cumulative variance

| variance | | | | | | |
|----------------------|---------------|--------|--------|--------|--------|--|
| PC | PC1 | PC2 | PC3 | PC4 | PC5 | |
| Eigenvalue | 200.39 | 104.57 | 71.88 | 52.76 | 32.47 | |
| % variance | 38.57 | 20.13 | 13.84 | 10.16 | 6.25 | |
| cumulative %variance | 38.57 | 58.70 | 72.53 | 82.68 | 88.94 | |
| Trait of interest | Eigen vectors | | | | | |
| 50%h | 0.008 | -0.068 | -0.125 | -0.085 | -0.051 | |
| no-spk/p | -0.019 | -0.020 | 0.017 | 0.002 | -0.020 | |
| FLA | 0.191 | 0.265 | 0.210 | 0.035 | -0.284 | |
| SL | 0.066 | 0.048 | 0.061 | -0.015 | 0.074 | |
| G/S | 0.738 | 0.315 | -0.524 | 0.176 | -0.089 | |
| SPKLT/SPK | 0.108 | 0.061 | 0.012 | -0.034 | 0.015 | |
| DTM | 0.019 | -0.042 | -0.063 | -0.094 | 0.032 | |
| PL | 0.035 | -0.129 | -0.051 | -0.136 | 0.301 | |
| PH | 0.438 | 0.093 | 0.423 | -0.686 | 0.301 | |
| BY | 0.298 | -0.215 | 0.420 | 0.597 | 0.384 | |
| PY | 0.036 | 0.166 | 0.230 | 0.315 | 0.210 | |
| 1000GW | 0.148 | -0.039 | 0.476 | 0.044 | -0.712 | |
| HI | -0.312 | 0.847 | 0.141 | 0.063 | 0.150 | |

analyzed with flag leaf area, plant height and biological yield. Harvest index was positively significantly associated

with plant yield while revealed a negative but significant relation with peduncle length and biological yield.

CONCLUSIONS

In present research it has been investigated that genetic diversity calculated through quantitative traits by multivariate analysis has importance in selection of genotypes for breeding programs. 1056 (sdf5) was recognized as more diversified genotype by PCA analysis with least days to maturity, highest FLA, plant yield and biological yield. AOC-YR/ATTILA (CGSS97Y0061S-32Y-1B-1B) (sdf30), AOC-YR/ATTILA (CGSS97Y0061S-142Y-1B-1B) (sdf27), 1076 (sdf9), 1072 (sdf6) and LALBMONO4*4/PRL//LALB (CGSS99Y00093S-2F1-22Y-9GHB-1GHB) (sdf32) are also superior in more than two morphological traits.

Recommendations

The resultant morphological superior genotypes are suggested to be screened for resistance genes and used in breeding programs. Correlation analysis showed positively and significantly association of plant yield with flag leaf

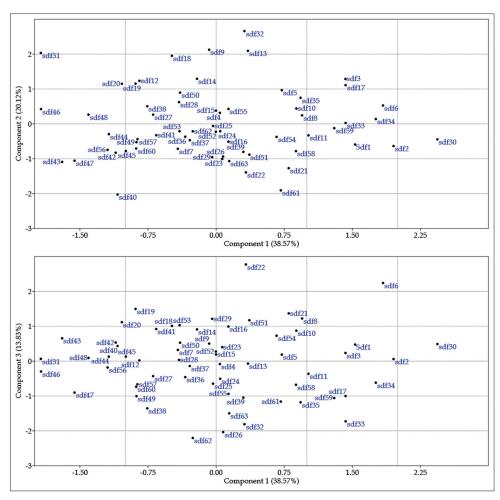


Fig 5. 2D scatter plot expressing genetic association among 63 wheat genotypes in first three PCs.

area, spike length and biological yield, these parameters should be major selection criterion for breeding studies.

Authors' contribution:

SB and IU designed the study. SB carried out the experiment and wrote the manuscript with support from AQ, KB and K. SB, AB and K did the statistical analysis. IU conceived the original idea and supervised the project.

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