Multivariate analysis and role of direct–indirect effect for yield and its component traits in bread wheat (*Triticum aestivum* L.)

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Abstract
The present study was carried out in a randomized block design with twenty-three diverse wheat (*Triticum aestivum* L.) genotypes. The experiment was conducted at Lok Bharti Wheat Research Centre, Bhavnagar Gujarat. Diversity analysis was done through D² and principal component analysis and the role of direct-indirect effect for yield and its component traits were estimated. Grain yield is influenced by the heading days, the number of tillers per meter, the number of grains per spike, thousand kernel weight and harvest index thus, selection of these traits would be effective. The highest inter-cluster distance was observed between clusters XI and X thus, genotypes that fall into these clusters showed wide genetic diversity and could be utilized in hybridization. The first three principal components accounted for 82.0 % of the total variability. The information obtained from this study is useful for parent selection in future wheat hybridization.

Keywords: Wheat, Diversity, Path coefficient and Principal component

INTRODUCTION
Wheat is the second most important cereal crop in the world followed by rice. The total area under wheat crop in India is 31.76 million hectares with a production of 109.52 million tones (Anonymous, 2021). Wheat is a self-pollinated crop that belongs to the genus *Triticum* and the family Poaceae. It has a relatively high content of niacin and thiamin which are principally concerned with providing the special protein called ‘Gluten’. Wheat proteins had special significance because gluten provides the framework for the spongy cellular texture of bread and baked products. India enabled record production of wheat in the last few years and attains the second-largest wheat producer globally. Wheat is very sensitive to biotic and abiotic stress, therefore, resulting in low productivity under the scenario of climate change. Genetic diversity present in the germplasm accessions is an important tool for any plant breeding program (Azam *et al.*, 2018).

The narrow genetic background has rendered improved varieties less tolerant to biotic and abiotic stresses (Maqbool *et al.*, 2010). There is a great scope to increase wheat production in timely sown conditions by breeding more efficient plant types, adaptable to variable environmental conditions. To improve the genetic contents of any crop, genetic variability is a prerequisite. Identification of potential genotypes and related attributes on variability is an important step in wheat breeding. The development of high-yielding wheat cultivars is a major objective in wheat breeding programs (Ehdaie and Waines, 1989). Yield is a complex trait, is a function of several component traits and their interaction with the environment (Khairnar *et al.*, 2018). In this regard path coefficient analysis helps in the partitioning of correlation coefficients into direct and indirect effects and in the assessment of the relative contribution of each component character.
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to the yield. Multivariate analysis, such as cluster analysis and principal component analysis (PCA) is the statistical procedure used to create the cluster in order to classify and identify divergent parents (Venkataramana et al., 2022). It is true that the more diverse the genetic base, the greater the chance of getting productive recombinants and broad variability in segregating generations (Mohammadi and Prasanna, 2003). Several methods for diversity analysis such as clustering, $D^2$, and principal component analysis were used in wheat breeding. Mahalanobis’s generalized distance is estimated by $D^2$ statistics for discriminating populations considering a set of parameters together rather than inferring from indices based on morphological similarities and polygenic relationships (Sankar et al., 2014; Rasitha et al., 2020; Swamynatham et al., 2020). The knowledge of the nature and magnitude of genetic variability for a heritable trait is essential to exploit the yield contributing trait. Assessment of genetic diversity distance via $D^2$ analysis, between two genotypes, will help to choose better parents for hybridization (Ghazy et al., 2015; Kiprotich et al., 2015; Sharma et al., 2020). Intercrossing between genetically divergent genotypes is expected to produce superior hybrids and desirable recombinants. The selection of parents based on the extent of genetic divergence has been successfully utilized in different crop species (Singh and Gupta, 1979; Jatasra and Paroda, 1978). The PCA-bi-plot is one of the utmost multivariate analyses to estimate the trait’s communication and genotypic presentation and is extensively used to separate the traits association (Aslam et al., 2017). It is the data reduction technique applicable to the quantitative types of data and transforms multi-correlated variables into another set of uncorrelated variables (Kumar et al., 2020).

**MATERIALS AND METHODS**

The experimental material consisted of 18 advanced entries and 5 released varieties (Table 1). The experiment was carried out during the rabi season of 2017-18 at Lok Bharti Wheat Research Centre, Sanosara, Bhavnagar, Saurashtra region of Gujarat. Geographically, Sanosara is located at 23.00N latitude, 072.00E longitude, and 67-meter elevation from mean sea level (MSL). The experiment was planted in Randomized Block Design (RBD) with three replications. Six rows were planted for each genotype and recommended agronomic package and practices were followed to raise the crop. Data were recorded on the five plants per replication per genotype. Data were recorded for the ten characters. Heading days were calculated from the date of sowing to 75% ear emergence. The number of effective tillers per meter was counted manually at the maturity stage. Spike length was taken in centimeters from primary spikes at maturity stages. Plant height was measured in centimeters from the above soil surface to the top of the plant excluding awns.

| S. No. | Variety | Parentage |
|-------|---------|-----------|
| 1     | VA 2016-02 | KLP 306 / PBW 573 |
| 2     | VA 2016-08 | GW 273 / MP 4010 |
| 3     | VA 2016-09 | GW 388 / MP 4010 |
| 4     | VA 2016-10 | PBW 550 / RAJ 4127 |
| 5     | VA 2016-11 | PBW 550 / RAJ 4127 |
| 6     | VA 2016-17 | WR 8849 / K 8236 / NIAW 1509 |
| 7     | VA 2016-20 | GW 336 / RAJ 4027 // GW 496 |
| 8     | VA 2016-25 | RAJ 4037 / KRL 13 // UP 2700 |
| 9     | VA 2016-32 | PBW 559 / WR 1873 |
| 10    | VA 2016-38 | DL327 / HI 1553 // GW 496 |
| 11    | J 16-11   | HW 3070/GW 496 |
| 12    | J 16-08   | KO 204/RAJ 4137//GW 2006-17 |
| 13    | J 16-01   | GW 273/GW 366 |
| 14    | J 16-14   | TOB/ERA/TOB/CNO-67/3//PLO/VEE 515/KAUZ/6//PBW 526 |
| 15    | J 16-24   | HPW 237/HI 1567 |
| 16    | J 16-15   | HW 2024/PHS 0721 |
| 17    | LOK 2017-3| PBW 588/CPAN 4061//HW 2006/HW 2002/S.S/C 306/S 331/HS 295/CPAN 4061/HW 2006/HW 2002//GUAM 92/BCN |
| 18    | LOK 2017-4| S.S/C 306/S 331/LOK 1//HW 888 |
| 19    | GW 496 (C)| HD 2285 / CPAN 1861 |
| 20    | GW 322 (C)| PBW 173 / GW 196 |
| 21    | GW 366 (C)| DL 802-3 / GW 232 |
| 22    | GW 451 (C)| GW324/4/CROC_1/AE.SQUARROSA (205)/JUP/ JY/5/ SKAUZ /4/ KAUV /5/ GW 339 |
| 23    | HI 1544 (C)| HIND162/BOBWHITE/CPAN2099 |

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average number of grains was calculated manually from random 10 well-developed spikes to observe the number of grains per spike. 1000 grain weight was estimated in grams with the help of weighing machines. Maturity Days were recorded when 75% of the plants show complete senescence. Threshed grains’ weight was recorded in kg per genotype per replication and quintal per hectare yield was calculated. Obtained data were subjected to the biometrical analysis that included path coefficient and multivariate analysis of Mahalanobis D². PCA statistics in the indoStat 8.1 version.

RESULTS AND DISCUSSION

Path coefficient analysis measures the direct influence of one variable upon the other and permits the separation of correlation coefficients into components of direct and indirect effects (Ali and Shakor, 2012). The phenotypic and genotypic path coefficients among the various characters are presented in Table 2. Path analysis explained that heading days and the number of grains per spike were the major contributors toward grain yield through the genotypic path while heading days and number of tillers per meter were major contributors through the phenotypic path. Genotypic path coefficient was observed that heading days showed the highest positive indirect effect on grain yield through the number of grains per spike, while the number of grains per spike had the highest positive indirect effect through heading days. On the other hand, phenotypic path coefficient was observed that heading days had the highest positive indirect effect on grain yield through the number of grains per spike and straw yield, while the number of tillers per meter had the highest positive indirect effect through spike length plant height, thousand kernel weight and maturity days. Therefore in the current study, selection for heading days, the number of tiller per meter and the number of grains per spike will be effective. In most instances, there was a close agreement between phenotypic and genetic path coefficient, while in the other case, the differences were high, signifying the importance of the environmental effect in estimating their parameter. Bergale et al. (2002) reported that tillers per plant had positive direct effects on grain yield. Dokuyucu and Akkaya, (1999) also reported a positive direct effect of the number of grains per spike and its positive association with grain yield. Aycicek and Yildirim, (2006) reported positive but small direct effects of thousand grain weight on grain yield in wheat. Mondal et al. (1997) obtained grains per spike, 1000-grain weight and tillers per plant had positive direct effects on grain yield.

The estimation of genetic divergence has been quantified as the relatedness or genetic distance between genotypes. Mahalanobis’s D² analysis employed for

### Table 2. Path coefficient with direct and indirect effect

| Characters                  | Phenotypic Direct / Genotypic effect | Number of plant/m | Heading days | Number of tiller/m | Spike length | Plant height | Number of grains/spike | 1000 grain weight | Maturity days | Straw yield | Correlation with grain yield |
|-----------------------------|-------------------------------------|-------------------|--------------|--------------------|--------------|--------------|------------------------|------------------|---------------|-------------|-------------------------------|
| Number of plants/m         | P                                   | -0.33             | -0.09        | 0.33               | 0.07         | 0.06         | -0.13                  | 0.01             | 0             | -0.02       | -0.11                         |
|                            | G                                   | 0.03              | -0.3         | 0.19               | 0.09         | 0.03         | -0.23                  | -0.02            | 0.01          | 0.01        | -0.15                         |
| Heading days               | P                                   | 0.6               | 0.05         | -0.08              | -0.07        | -0.08        | 0.17                   | -0.02            | -0.07         | 0.06        | 0.55**                        |
|                            | G                                   | 1.14              | -0.01        | -0.05              | -0.07        | -0.04        | 0.22                   | -0.03            | -0.45         | -0.04       | 0.67                          |
| Number of tiller/m         | P                                   | 0.41              | -0.26        | -0.12              | 0.07         | 0.06         | -0.14                  | 0.01             | 0.01          | -0.02       | 0.02                          |
|                            | G                                   | 0.2               | 0.03         | -0.3               | 0.1          | 0.04         | -0.23                  | 0.02             | 0.05          | 0.02        | -0.09                         |
| Spike length               | P                                   | -0.13             | 0.18         | 0.31               | -0.22        | -0.08        | 0.22                   | -0.04            | -0.02         | 0.04        | 0.26*                         |
|                            | G                                   | -0.12             | -0.02        | 0.66               | -0.16        | -0.04        | 0.29                   | -0.06            | -0.14         | -0.02       | 0.39                          |
| Plant height               | P                                   | -0.13             | 0.14         | 0.36               | -0.19        | -0.08        | 0.15                   | 0.03             | -0.05         | 0.06        | 0.28*                         |
|                            | G                                   | -0.06             | -0.02        | 0.72               | -0.13        | -0.08        | 0.2                    | 0.04             | -0.03         | 0.03        | 0.35                          |
| Number of grains/spike     | P                                   | 0.39              | 0.11         | 0.26               | -0.15        | -0.08        | -0.05                  | -0.11            | -0.02         | 0.02        | 0.37**                        |
|                            | G                                   | 0.48              | -0.01        | 0.51               | -0.1         | -0.08        | -0.02                  | -0.16            | -0.15         | -0.02       | 0.45                          |
| 1000 grain weight          | P                                   | 0.24              | -0.01        | -0.06              | 0.01         | 0.02         | -0.01                  | -0.18            | 0.01          | 0.01        | 0.02                          |
|                            | G                                   | 0.33              | 0            | -0.12               | 0.01         | 0.02         | -0.01                  | -0.24            | 0.05          | 0.01        | 0.04                          |
| Maturity days              | P                                   | -0.09             | 0            | 0.5                | -0.04        | -0.04        | -0.07                  | 0.11             | -0.02         | 0.05        | 0.41**                        |
|                            | G                                   | -0.49             | 0            | 1.03               | -0.02        | -0.04        | -0.03                  | 0.15             | -0.03         | -0.04       | 0.52                          |
| Straw yield                | P                                   | 0.1               | 0.06         | 0.36               | -0.07        | -0.05        | 0.09                   | 0.02             | -0.05         | 0.05        | 0.40***                       |
|                            | G                                   | -0.04             | -0.01        | 1.11               | -0.09        | -0.07        | 0.19                   | 0.06             | -0.47         | 0.62        |                               |

Residual effect = 0.437
the grouping of 23 wheat genotypes using 10 yield and yield attributing traits resulted in eleven clusters. Cluster I consist of a maximum number of genotypes, while the remaining clusters had a single genotype (Table 3). Inter and intra-cluster distances were measured to estimate the level of diversity among the genotypes. Inter and intra-cluster distances were estimated and presented in Table 4. The maximum intra-cluster distance was reported for cluster I. Whereas, the maximum inter-cluster distance was observed between clusters X and XI, followed by clusters V and XI, clusters VII and IX. On the other hand, the minimum inter-cluster distance was observed between clusters VIII and VI. The Maximum inter-cluster distance suggested wide diversity between the genotypes, while the minimum inter-cluster distance indicated a close relationship (Waiker and Arun, 2015). Dobariya et al. (2006) noticed that genotypes within clusters with a high degree of divergence would produce more desirable breeding materials for attaining the maximum genetic advance. The higher inter-cluster distance than intra-cluster distance showed homogeneity and narrow genetic variability within a cluster (Singh and Gupta, 1979; Malik et al., 2017). The maximum diversity for all studied traits was observed between VA 2016-09 (cluster X) and J 16-08 (cluster XI). The pedigree of VA 2016-09 (GW 388 / MP 4010) and J 16-08 (KO 204/RAJ 4137//GW 2006-17) also differed significantly. Therefore the hybridization between VA 2016-09 and J 16-08 exploits maximum heterosis. More intra-cluster distance may be due to the degree of heterogeneity and pedigree and hence, selection will be efficient if it is based on the highest mean for desirable traits (Ramya et al., 2017; Kaushik et al., 2018; Rasitha et al., 2020). Dobariya et al. (2006) noticed that genotypes within clusters with a high degree of divergence would produce more desirable breeding materials for attaining the maximum genetic advance. The higher inter-cluster distance than intra-cluster distance showed homogeneity and narrow genetic variability within a cluster (Singh and Gupta, 1979; Malik et al., 2017). The maximum diversity for all studied traits was observed between VA 2016-09 (cluster X) and J 16-08 (cluster XI). The pedigree of VA 2016-09 (GW 388 / MP 4010) and J 16-08 (KO 204/RAJ 4137//GW 2006-17) also differed significantly. Therefore the hybridization between VA 2016-09 and J 16-08 exploits maximum heterosis. More intra-cluster distance may be due to the degree of heterogeneity and pedigree and hence, selection will be efficient if it is based on the highest mean for desirable traits (Ramya et al., 2017; Kaushik et al., 2018; Rasitha et al., 2020). Maximum inter-cluster distance also leads to a wide spectrum of variability in segregating populations to operate selection (Singh et al., 1981; Govindaraj et al., 2011; Athoni et al., 2016). Cluster XI had the highest mean value for six traits days to heading, spike length, plant height, the number of grains per spike, maturity days, and straw yield (Table 5). Cluster IX had the highest mean value for grain yield, cluster V for thousand-grain weight, cluster IV for the number of plants per meter and the number of tillers per meter. The lowest mean values for different traits were also significant. Cluster II had the lowest mean

Table 3. Distribution of genotypes into eleven cluster based on D² value

| Cluster | I   | II  | III | IV  | V   | VI  | VII | VIII | IX  | X   | XI  |
|---------|-----|-----|-----|-----|-----|-----|-----|------|-----|-----|-----|
| I       | 30.2| 52.2| 48.8| 54.1| 69.9| 49.3| 95.4| 54.5 | 93.0| 84.6| 145.2|
| II      | 0.0 | 122.1| 55.6| 155.6| 68.2| 34.0| 74.4| 216.6| 119.3| 163.3|
| III     | 0.0 | 85.4| 28.4| 28.4| 86.1| 172.0| 125.9| 33.4 | 55.9| 239.3|
| V       | 0.0 | 109.8| 213.3| 135.9| 39.6| 58.7| 267.8|
| VI      | 0.0 | 107.1| 27.0| 115.4| 173.8| 63.2|
| VII     | 0.0 | 99.6| 254.5| 131.1| 223.1|
| VIII    | 0.0 | 139.8| 184.9| 46.9|
| IX      | 0.0 | 107.2| 258.0|
| X       | 0.0 | 384.0|
| XI      | 0.0 |      |

Table 4. Average inter and intra cluster distances for eleven clusters

| Cluster number | Number of varieties | Name of varieties in a cluster |
|----------------|---------------------|--------------------------------|
| I              | 13                  | J 16 15, GW 496, HI 1544, VA 2016-02, VA 2016-20, VA 2016-32, J 16-11, LOK 2017-03, J 16-01, VA 2016-11, VA 2016-10, GW 451, VA 2016-98 |
| II             | 1                   | VA 2016-17 |
| III            | 1                   | VA 2016-25 |
| IV             | 1                   | J 16-24    |
| V              | 1                   | GW 366     |
| VI             | 1                   | J 16 14    |
| VII            | 1                   | VA 2016-38 |
| VIII           | 1                   | GW 322     |
| IX             | 1                   | LOK 2017-04|
| X              | 1                   | VA 2016-09 |
| XI             | 1                   | J 16-08    |

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value for heading days, cluster VII for plant height and cluster X had the lowest mean value for maturity days. In future breeding programs, genotypes could be selected for higher spike length, the number of grains per spike and straw yield from cluster I. Whereas in the current study LOK 2017-04 had the highest mean value for grain yield therefore, it is considered as best performing entry, which falls under cluster IX. Similarly, for heavy rainfall areas, the genotypes may select from cluster VII because minimum plant height was observed and for the drought-prone area, the genotypes from clusters II and X could be selected due to early maturity.

The principal component analysis is a nominal approach for dropping the variability in multiple traits to the principal components with the first principal component arresting the maximum variability. Principal component analysis with a correlation matrix is the finest to control the principal factors, as it does not involve the normal distribution assumption of populations (Chaudhary et al., 2015; Sharma et al., 2020). PC with greater eigenvalues and variables with high factor loadings were considered the best representative of system attributes. In the present investigation, PC was performed for yield and other attributes in wheat (Table 6 and Fig. 1).

The maximum variability (48.0%) was reported for PC1, followed by PC2 (20.0%) and PC3 (14.0 %). The first three PC explain 82.60 % of the total variability. Similarly, Khan et al. (2015) revealed that the first three principal components explained 82.60% of the total variation.

![Fig.1. Scree plot of principal component show eigenvalue on Y axis and principal component on X axis](https://doi.org/10.37992/2022.1302.045)
Table 6. Vector loadings and percentage explained variation by the PCs

| Variables                  | PC1     | PC2     | PC3     |
|----------------------------|---------|---------|---------|
| Number of plant / m        | 0.27    | -0.50   | 0.12    |
| Heading days               | -0.39   | -0.30   | 0.02    |
| Number of tiller / m       | 0.30    | -0.48   | 0.16    |
| Spike length               | -0.37   | 0.26    | 0.05    |
| Plant height               | -0.38   | 0.03    | -0.29   |
| Number of grain / spike    | -0.31   | 0.15    | 0.48    |
| 1000 grain weight          | 0.06    | -0.07   | -0.77   |
| Maturity days              | -0.32   | -0.42   | -0.03   |
| Grain yield                | -0.28   | -0.27   | 0.15    |
| Straw yield                | -0.37   | -0.29   | -0.17   |
| Eigen values               | 4.76    | 2.03    | 1.35    |
| Proportion of variance (%) | 48.0    | 20.0    | 14.0    |
| Cumulative proportion of variance (%) | 48.0 | 68.0 | 81.0 |

(Ramya et al., 2017; Jain and Diwan, 2021) recommended considering the characters or genotypes showing more PC1 scores for catching the variability of a particular trait. Based on factor loading of PCA analysis PC1 was highly related to the number of tillers per meter, followed by the number of plants per meter and thousand-grain weight. The PC2 was highly related to spike length, followed by the number of grains per spike and plant height, while PC3 was highly related to the number of grains per spike followed by the number of tiller per meter, grain yield and the number of plants per meter.

The present study illustrated the existence of a wide range of variations for most of the traits among wheat genotypes and opportunities for genetic gain through selection or hybridization. Hence, improving one or more traits could result in a high grain yield for hexaploid wheat. Heading days, the number of grain per spike, thousand-grain weight, the number of tiller per meter and straw yield had positive phenotypic direct effect therefore selection of these traits improves grain yield positively. The distribution pattern of all the genotypes into various clusters showed the presence of considerable genetic divergence among the genotypes for most of the traits. Wheat genotypes were grouped into eleven clusters. The highest inter-cluster values were observed between clusters XI and X thus genotypes included in these clusters showed wide genetic diversity hence these genotypes may be selected as parents in hybridization programs to exploit maximum heterosis.

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