

Biplot analysis of seed yield and oil content combining ability in rapeseed (*Brassica napus* L.)

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Abstract

Biplot analysis was conducted to estimate the combining ability of ten parental genotypes using 7×3: Line×Tester data of 21 F₁ hybrids in (*Brassica napus* L.) for seed yield and oil content. The data obtained from all the three replications of experimental material in randomized complete block design were subjected to analysis of variance to check the null hypothesis of the equality of means. Significant differences (P≤0.01) were found among the parents, F₁ hybrids and Line×Tester interaction for the variables seed yield plant⁻¹ and oil content. The genotypes CA-4 and DH-6 were the promising for seed yield plant⁻¹ (36.8 g) and oil content (51.2%), respectively. Among the hybrids DH-3×CA-4 and DH-4×CA-4 were best for seed yield plant⁻¹ (51.1 g) and oil content (52.1%). The GGE biplot displayed DH-8 and DH-2 as the best general combiners for seed yield and oil content, respectively. The best specific combiner for seed yield plant⁻¹ was DH-3 whereas; for oil content DH-2 was ranked first. Among the testers CA-5 and CA-4 were ranked first on the basis of GCA for seed yield plant⁻¹ and oil content, respectively. The GGE biplot graphic allowed a rapid and effective overview of General Combining Ability (GCA) and Specific Combining Ability (SCA) effects of the inbred lines; best lines and tester, as well as their performance in crosses.

Keywords: *Brassica napus* L.; GGE biplot; GCA; line x tester and SCA.

Abbreviations: GCA_general combining ability; SCA_specific combining ability.

Introduction

Worldwide *Brassica* species are the third most important source of oils and their production has viewed a steady rising movement through modern and conventional plant breeding approaches (Beckman, 2005). The rapeseed breeding programs are mainly aimed to produce varieties with high yield amount of grain and oil per unit area. Currently, although the planted cultivars are capable of achieving high oil content, but further improvement of grain yield and percentage of oil through different breeding activities is required. *Brassica* breeding activities are chiefly aimed with the objectives to produce new genotypes with constant and high grain yield and oil percentage as well as fewer amounts of glucosinolate and erucic acid contents. Grain yield and oil content are quantitative traits influenced by the genotype, environment and interaction between these two (Huhn and Leon, 1985). The complex nature of these traits happens due to the diverse processes that take place during plant development stages. The utility of genetic variability in any crop species is considered to be critical for making further genetic progress in grain yield as well as other economically important traits (Rehman et al., 2009). Development of superior varieties to enhance the production and yield has always been one of the major strategies in agricultural development programs.

Reshuffling the genes through hybridization from proper parents could produce superior varieties. Moreover, it is also

necessary to know about the nature and magnitude of gene action responsible for controlling the inheritance of various yield attributes along with combining ability of the parents and their cross combinations to use in further crop improvement programs. In rapeseed breeding program for hybrid and open pollinated varieties, general and specific combining ability effects (GCA and SCA) are important indicators of the potential of inbred lines in hybrid combinations. The line × tester analysis is one of the efficient methods of evaluating large number of inbred as well as providing information on the relative importance of GCA and SCA effects for interpreting the genetic basis of important plant traits (Singh and Chaudhury, 1977).

Some methods are available to analyze diallel crosses, such as the most popular method of Griffing. Griffing (1956) partitioned the total variance to GCA variance of parents and SCA variance of crosses. Yan and Hunt (2002) suggested a new approach of the principal component biplot technique for diallel analysis. This technique enhances the capability of interpreting the phenotypic variation to obtain combining ability and inter-relationships of parents based on graphical presentation using PC1 and PC2, which are derived through PC analysis of environment-centered yield data. Thus, combining ability analysis using GGE biplot is needed to visualize the GCA and SCA of each genotype, groups of parents with similar genetics and superior hybrids. Similarly,

in multilocation trials, the first two components can be used to visualize GCA and SCA effects (Bertoia et al., 2006).

The GGE biplot analysis of diallel data is widely used to determine combining ability and heterotic responses in many crops and many traits, such as in wheat (Dehghani et al., 2013; Malla et al., 2010), melon (Dehghani et al., 2012), linum (Rastogi et al., 2011), opium (Rastogi et al., 2013), cotton (Hamoud, 2014) and maize (Mostafavi et al., 2012; Badu-Apraku et al., 2013). This method is rarely reported for line \times tester data analysis. Fotokian and Agahi (2014) reported that identification of suitable parents, heterotic crosses and the best hybrids in line \times tester data using GGE biplot.

Keeping in view the utility of combining ability and heterosis in plant breeding, the present study was planned with the objectives to estimate GCA and SCA of the parents, to identify the heterotic groups and to formulate hypotheses on the genetic relations of the parents.

Results and Discussion

Seed yield plant⁻¹

Genotypes showed significant differences ($P \leq 0.01$) for seed yield plant⁻¹. Also the differences observed among the parents, lines, testers, the F₁ hybrids and interaction of (L \times T) were defined as significant ($P \leq 0.01$) by the analysis of variance. The contrast comparison of both L vs T and P vs F₁ was significant ($P \leq 0.05$) (Table 1). Seed yield is the final outcome resulting from various components. Almost in all the crops that are grown for their utilizable seeds, genotypes with higher yield of the seed are desirable. Our analysis of the seed yield data exposed high variation among the parents and hybrids that may lead towards the effective isolation of desirable observations. These results are also supported by the findings of Khan et al. (2006) and Sabaghnia et al. (2011).

The parental mean for seed yield ranged from 20.1g and 36.8g for DH-6 and CA-4, respectively. The hybrid DH-3 \times CA-4 showed the best mean seed yield over the other DH-2 \times CA-2 hybrid and DH-8 \times CA-5. It can be inferred that the heterotic expressions exhibited by these F₁ experimental hybrids may be due to complementation among the loci related to grain yield per plant (Fig 1).

The absence of heterosis in most crosses for grain yield per plant can be directly related to its mode of inheritance. Amiri-Oghan et al. (2009) observed that this variable is controlled mainly by additive effects. However, according to Xing et al. (2014), grain yield per plant is mainly controlled by the additive and epistatic effects. In this context, it can be inferred that the absence of heterosis is not decisive absence of dominance. The negative heterosis in the hybrids performance compared to their parents must be justified by the dominance directed to the reduction of trait expression.

In the event of loci with positive and negative dominance, the effects can be annulled and heterosis not expressed. The dominance of interactions and/or over dominance not always indicate the direction of increasing the phenotypic value of the character. It should be emphasized that the occurrence of the interactions, in hybrids will constitute the best strategy to be adopted for the improvement and not the selection of superior individuals (Ramalho et al., 2012).

The biplot analysis for seed yield combining ability explained 86.5 % of the entire variation, out of which 60.8 % was specified for PC₁ and 25.7 % for PC₂. The parental line DH-8 was the best general combiner among the lines, while

DH-7 was placed as the best specific combiner. However, DH-8 interacted comparatively more with the testers; therefore, it can also be concluded as the best for SCA. Among the testers, CA-5 was placed as the best general combiner because of its most discriminating and representative ability (Fig 3B). Both the general and specific combining ability effects of the lines were significant. This significance of the GCA and SCA effects suggest the importance of both additive and non-additive gene actions for seed yield. Which were also reported the same by Panhwar et al. (2008) in line \times tester crosses of cotton. The effect of GCA of the testers was also significant because of their placement into different sectors. Two heterotic groups i.e. [DH-7, DH-2] \times [CA-2] and [DH-3, DH-5] \times [CA-4] were identified. Out of these, DH-3 \times CA-4 was the best hybrid combination for seed yield plant⁻¹. DH-8 showed a better average performance across all the testers but the best specific hybrids was not observed in any of the heterotic groups. This confirms its ability as best general combiner but the best hybrids were produced by some other parents (Fig 3A). The same was observed in some other testers, whereas the best general combiner did not fall into any of the heterotic groups. Therefore, they can be used as parents when aiming at breeding synthetic varieties, while for having vigorous hybrids the parents presented in the heterotic groups should be chosen. Sabaghnia et al. (2011) also reported the presence of sufficient heterosis in individual crosses but the parents in those crosses were not presented as the best general combiner.

Oil content

Significant differences ($P \leq 0.01$) were shown by the analysis of variance among genotypes, parents, lines, the F₁ hybrids and interaction (L \times T) for oil content. The differences among the testers were significant ($P \leq 0.05$), while non-significant ($P > 0.05$) for both of the contrasts i.e. P vs F₁ and L vs T (Table 1). In any oil seed crop breeding program one of the major objectives is the improvement of oil content level of the seed. Therefore, significant differences among the parents and hybrids are attractive for seed oil breeders. Our studied genotypes explored enough potential for selection towards improvement. Khan et al. (2006) also reported similarly significant genotypic effect for oil content. The parental genotype DH-6 yielded maximum oil content (51.5 %), while minimum oil content (45.8 %) was observed for DH-3. Among the F₁ hybrids DH-4 \times CA-4 and DH-2 \times CA-5 were the best combinations equally with maximum oil content (52.1 %), while minimum oil content (44.6 %) was produced by both of the cross combinations DH-3 \times CA-2 and DH-6 \times CA-5 (Fig 2).

Biplot analysis of oil content combining ability explained a sum of 94.6 % of the total variation. Out of which, 63.6 % is counted for PC₁ and the rest 31 % for PC₂. Maximum general combining ability was observed for the parental line DH-2 followed by DH-7, while minimum for DH-5 and DH-6. For specific combining ability DH-7 was identified as the best followed by DH-2. Both the GCA and the SCA effects of the lines were significant because of the considerable differences between their projections onto the ATC abscissa and ordinate. Among the testers CA-4 was identified as the best general combiner, as it displayed the most discriminating ability and representativeness (Fig 4B). The GCA effect of testers was also significant because of their settlement into different sectors. The polygon view of the biplot defined two heterotic groups i.e. [DH-7] \times [CA-2, CA-4] and [DH-2, DH-

Table 1. Analysis of variance table for seed yield plant⁻¹ and oil content.

SOV	DF	Seed yield plant ⁻¹		Oil content	
		MS	P-Value	MS	P-Value
Replication	2	9.99	0.00	0.25	0.41
Genotypes	30	191.66	0.00	12.71	0.00
Parents	9	81.21	0.00	8.75	0.00
Lines (L)	6	43.14	0.00	9.24	0.00
Testers (T)	2	58.4	0.00	2.26	0.03
L vs T	1	355.2	0.05	18.82	0.09
F ₁	20	190.99	0.00	15.1	0.00
Lines (L)	6	341.96	0.00	16.7	0.00
Testers (T)	2	8.35	0.02	49.85	0.00
L x T	12	145.94	0.00	8.51	0.00
P vs F ₁	1	1199.11	0.03	0.48	0.54
Error	60	1.50		0.27	
CV %		3.03		1.03	

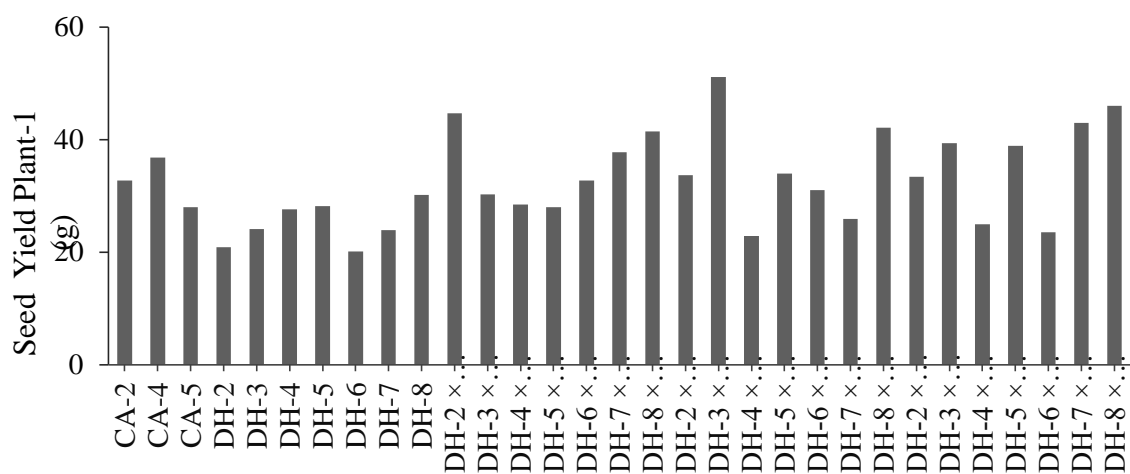


Fig 1. Mean performance of parental genotypes and F₁ hybrids for seed yield plant⁻¹.

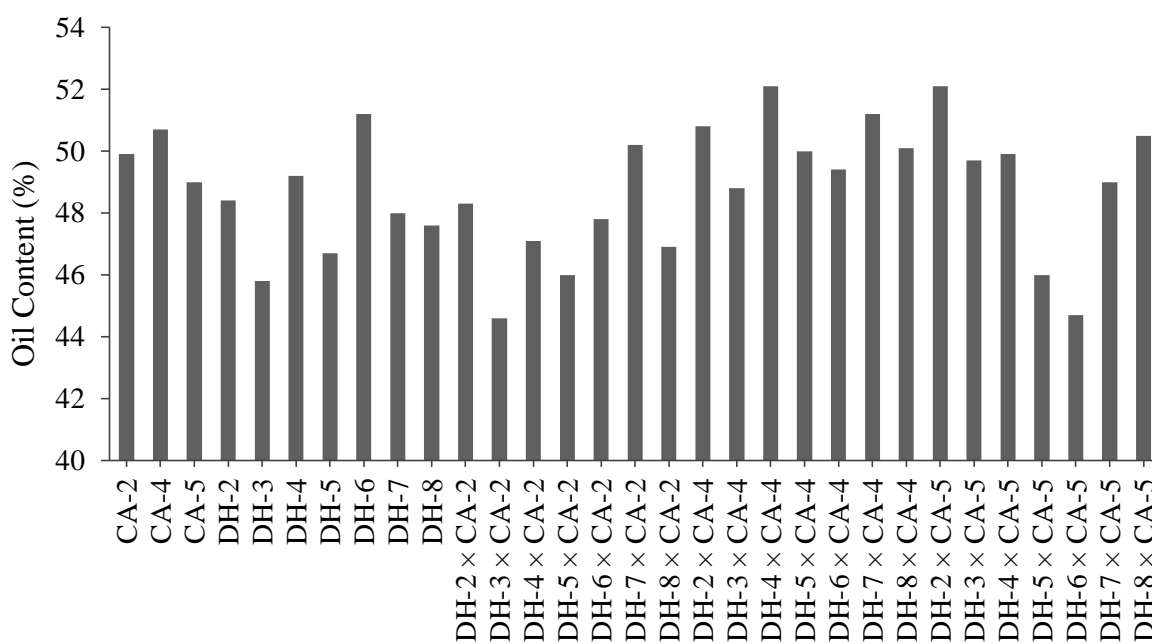


Fig 2. Mean performance of parental genotypes and F₁ hybrids for oil content.

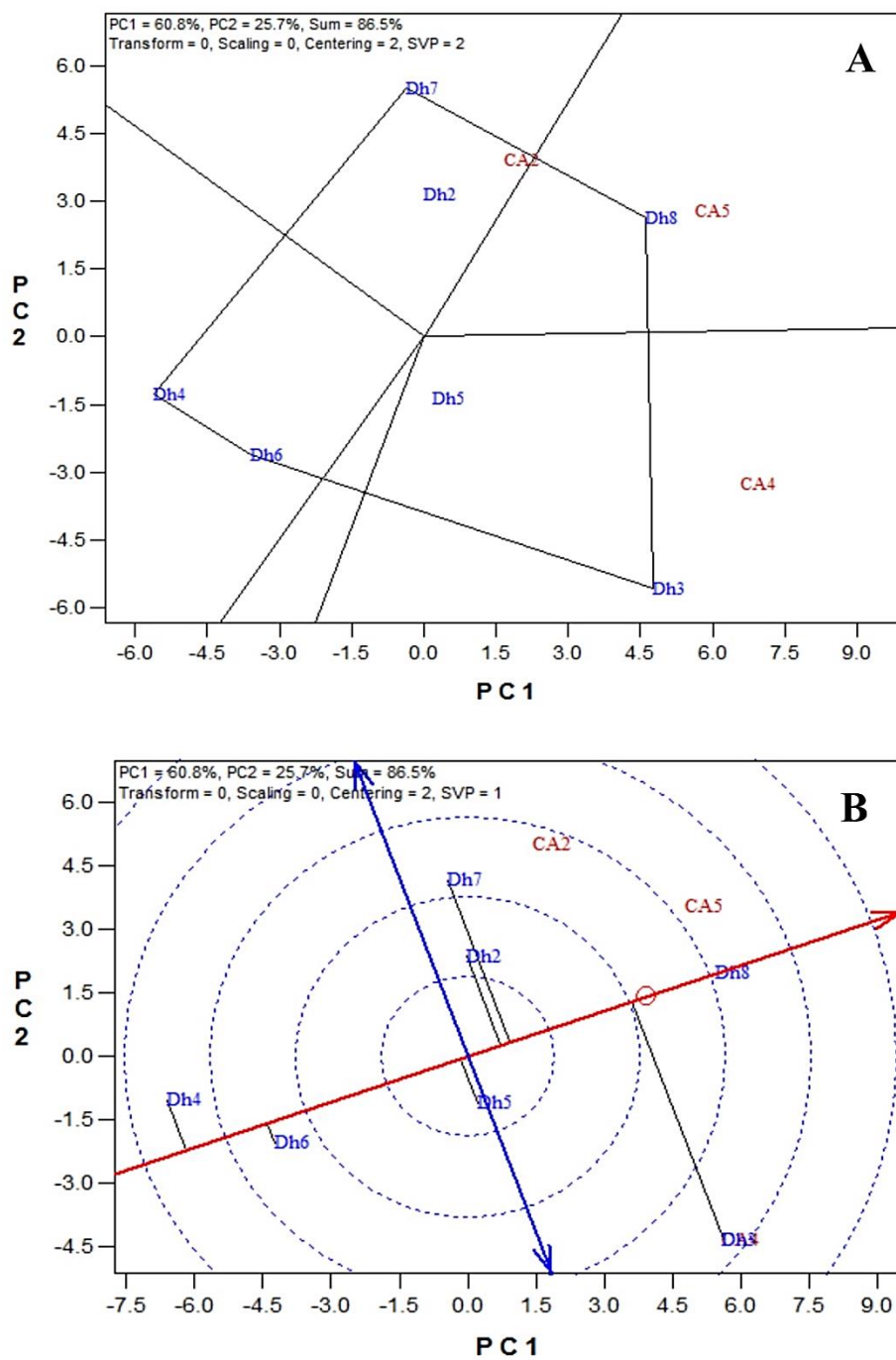


Fig 3. Biplot base on line × tester data in *Brassica napus* for seed yield plant⁻¹ (A) Polygon view for best hybrid combinations, (B) Average tester coordination view for GCA and SCA.

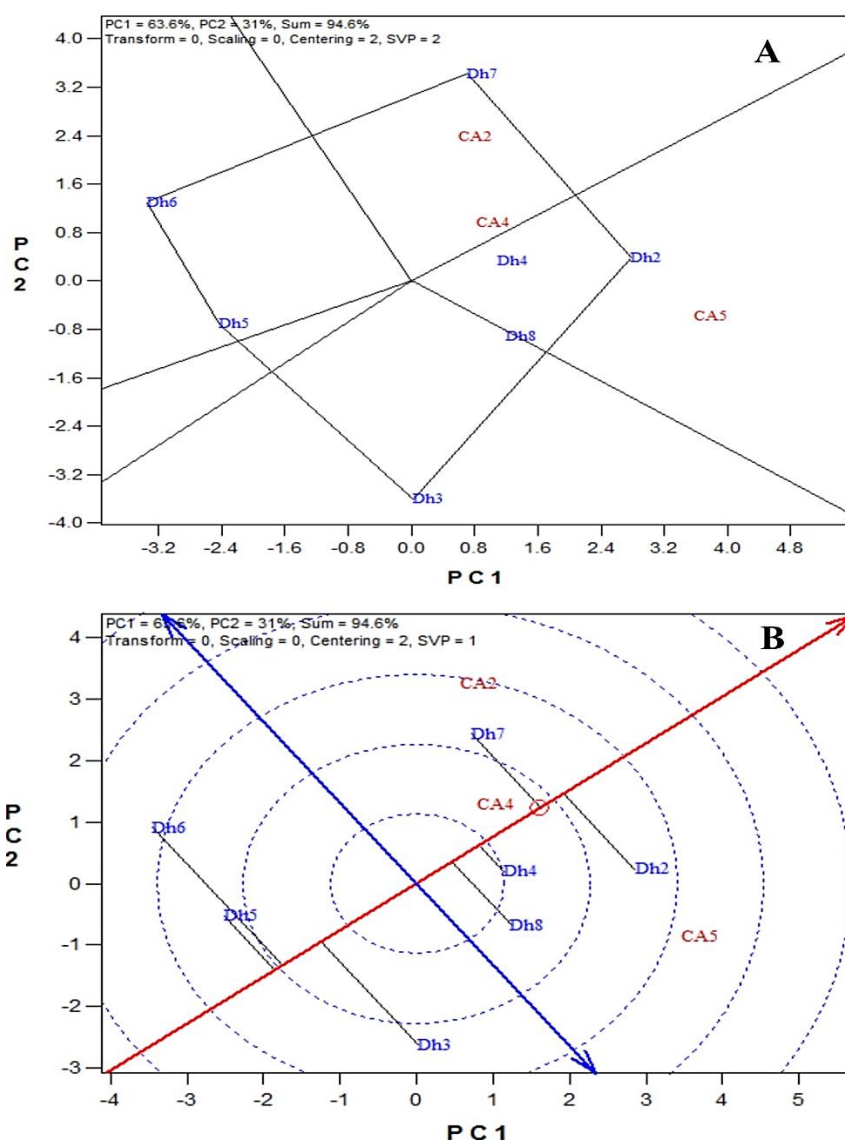


Fig 4. Biplot based on line \times tester data in *Brassica napus* for oil content; (A) Polygon view for best hybrid combinations, (B) Average tester coordination view for GCA and SCA.

4, DH-8] \times [CA-5, CA-4], in which DH-2 \times CA-5 and DH-4 \times CA-4 were identified as the best hybrids for oil content. Sabaghnia et al. (2010) also identified two heterotic groups from the biplot for oil content in half diallel of nine rapeseed cultivars and recommended those between group crosses comparatively better and also some of them as better from both the parents. According to our findings DH-6 was the best among the parents but is not presented in any of the cross; therefore; it was also explored as the worst general combiner for oil content (Fig 4A). Aher et al. (2009) also reported that the crosses with higher parents *per se* performance may also involve the parents with poor phenotypes for that specific trait.

Materials and Methods

Plant material and experimental design

A set of ten (*Brassica napus* L.) genotypes including seven double haploid Chinese lines and three local cultivars was used to produce F_1 hybrids by crossing them in a line \times tester

mating fashion. Twenty one F_1 progenies produced during crop season 2010-2011 were evaluated along with ten parental genotypes in randomized complete block design with three replications during cropping season 2011-2012. All the genotypes were grown, each in 3 rows of 5 meter length, each. Plant spacing was maintained as 50 cm and 30 cm between rows and within rows between plants, respectively. Standard cultural practices were adapted for the entire experiment.

Statistical analysis

The data collected on seed yield plant⁻¹ and oil content were subjected to analysis of variance. The significant Lines \times Testers interaction (L \times T) effect for both of the variables were analyzed for combining ability of the parents using GGE biplot methodology. Applying GGE biplot methodology to line \times tester data, the terms "average yield" and "stability" of the genotypes correspond to GCA and SCA, respectively. During GGE biplot analysis, means of each column are calculated and a new. The adjusted (non-symmetrical) data

matrix is obtained by subtracting the column (tester) mean from each cell. In this matrix, each population corresponds to one row and one column, where the row is considered as “line” and the column as “tester” (Yan and Hunt, 2002). After obtaining the first two principal components of the adjusted data matrix, the biplot model became as:

$$Y_{ij} - \beta_j = \lambda_1 \xi_{i1} \eta_{j1} + \lambda_2 \xi_{i2} \eta_{j2} + \varepsilon_{ij}$$

Where, Y_{ij} is the genotypic value of the cross between entry i and tester j for the trait of interest; β_j is the mean of all crosses involving tester j ; λ_1 and λ_2 are the singular values for PC1 and PC2, respectively; ξ_{i1} and ξ_{i2} are the PC1 and PC2 eigenvectors, respectively. For entry i ; η_{j1} and η_{j2} are the PC1 and PC2 eigenvectors, respectively, for tester j ; and ε_{ij} is the residual of the model associated with the combination of entry i and tester j . Principal components scores for entries and testers were scaled symmetrically (Yan and Hunt, 2002; Bertoia et al., 2006). The analyses were conducted using the GGE-biplot software; a window based application that fully automates biplot of two-way data (Yan, 2001).

Conclusions

The GGE biplot graphic allowed a rapid and effective overview of General Combining Ability (GCA) and Specific Combining Ability (SCA) effects of the inbred lines, best lines and tester, as well as their performance in crosses. The genetic variability among genotypes, allows the exploitation of additive and non-additive effects for seed yield plant⁻¹ and oil content. The cross combination DH-3×CA-4 was best for seed yield, whereas DH-4×CA-4 and DH-2×CA-5 equally best for oil content. Based on average tester coordination function of GGE biplot, the parental genotypes DH-8 and CA-5 were identified as the best combiner for seed yield plant⁻¹, whereas for oil content DH-2 and CA-4 were the best combiner parents. Further breeding strategies could be planned to exploit the potential of CA-4 for both seed yield and oil content. Among the lines DH-8 and DH-2 could be further utilized in breeding programs.

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