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Research Article

Study on gene action, combining ability and heterosis for different traits in Indian mustard (*Brassica juncea* **L. Cxern & Coss)**

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Abstract

Combining ability and heterosis was investigated in mustard hybrids obtained from 7 × 7 half diallel cross. Twenty one F_{1s} along with seven parents were evaluated in Randomized Complete Block Design. The combined analysis of variance revealed very significant differences among the parents for all morphological variables with the exception of secondary branches and siliqua length, indicating wide diversity among the parental material used in the present study. Significant GCA and SCA variance indicated additive and non-additive gene action across all the characters. The SCA variance components were larger than the GCA variance components for most traits indicating the prevalence of nonadditive gene effects. Jawahar mustard × PM-30, showed a significantly positive SCA and better parent heterosis for seed yield per plant whereas five hybrids (Kranti × PM-30, Gujarat Mustard-3 × Pusa Mahak, Jawahar Mustard × PM-30, Jawahar Mustard × Pusa Mahak and PM-30 × Pusa Mahak) exhibited significant positive SCA effect for number of siliqua/ plant. Jawahar Mustard × PM-30, Varuna×Jawahar Mustard, Gujarat Mustard-3 × PM-30, exhibited highly significant heterosis over the mid-parent.

Keywords: *Brassica juncea*, GCA, SCA, gene action, Heterosis

INTRODUCTION

Mustard [Brassica juncea L. (Czern&coss)] is India's most important oil seed crop. India's mustard production is much lower than the global average (Rana *et al.,* 2021). Increasing mustard production depends on cultivating more acreage or introducing new high-yielding varieties. Since space constraints restrict area expansion, development of new mustard varieties with high genetic potential for crop yield is the favourable option. The diversity of plant types and ease of crossing, in conjunction with the crop's high adaptability, presents a good opportunity for enhancing yield *via* recombination breeding, transgressive segregant selection, and heterosis breeding in mustard (Singh *et al.,* 2022; Lakshman *et al.,* 2020). Plant breeding is governed by the genetic information available from parents and their cross combinations (Singh *et al.,* 2016; Sandhu *et al.,* 2019). To start a successful mustard breeding strategy, it is essential to identify the genetic type and the estimated pre-potency of parents in hybrid combinations.

Selecting parents from several heterotic groups has the potential to increase hybrid vigour (Lakshman *et al.,* 2018; Singh *et al.,* 2022). To generate such genetic information from parents and progenies, as well as their general and specific combining ability, a genetic model in respect to the experimental material is required. A variety of models have been developed to predict the general and specific combining ability of parents and crosses. Diallel analysis is one of these methods that is useful for estimating genetic parameters and providing information on the genetic behaviour of the traits under study. This technique has been employed in a variety of crops, including mustard (Kaur *et al.,* 2022; Tele *et al.,* 2016; Gupta *et al.,*

2010). Griffing (1956 a, b) classified diallel crosses into four categories. Of the four diallel approaches, half diallel procedure (without reciprocal crosses) have several advantages over others, providing the most information about the genetic architecture of a trait, parents, and allelic frequency, and are the most commonly used due to their ease of use. The combining ability in this context explains the breeding values of parental lines in order to develop specified cross combinations. Crossing a line to various others provides the mean performance of the line in all its crosses. This mean performance, represented as a divergence from the mean of all crosses, is referred to as a line's general combining ability (GCA). The predicted value of every individual cross is thus the sum of the general combining abilities of its two parental lines. The cross, on the other hand, may diverge from this expected value to a greater or smaller extent. This difference is referred to as the specific combining ability (SCA) of the two lines in combination. The GCA effects aid in the selection of superior parents, while the specific combining ability effects aid in the selection of superior hybrids. Crop breeding programs that include at least one parent with a high GCA value and a large SCA impact, as well as a hybrid with high *per se* performance are more dependable than those that do not include at least one parent with a high GCA value when making parent selections (Fasahat *et al.,* 2016). Several heterosis values for grain production in mustard have been reported by researchers.

MATERIALS AND METHODS

The parental lines for the study were obtained from the Pulses and Oilseeds Research Station in Berhampur, West Bengal, India. Seven lines /genotypes were selected as parents based on evaluation of diverse agro-morphological traits during *Rabi* season 2018- 2019 (October-February) (**Table 1**). Hybridization was started at the onset of flowering during *Rabi* season 2018-2019 (October-February) among the parents based on flowering synchrony. Emasculation was done in the afternoon (3 pm to 6.30 pm). Only those flower buds, which were expected to open in the next morning, were chosen for emasculation.

Twenty-one F_{1s} and their seven parents were evaluated in randomised complete block design (RCBD) with three

Table 1. **List of parental materials used in Experiment**

replications with a plot size of 3×2 m² having spacing 60 cm × 20 cm. during *Rabi* season 2019-2020 (October-February) at the Agricultural Farm, Institute of Agriculture, Visva-Bharati, Sriniketan, located at 23° 19' N latitude, 87° 42' E longitude, and 58.9 m above sea level using a 7×7 half-diallel mating design. All the recommended package of practices were adopted to grow a good crop. Standard hybridization techniques (Labana *et al.,* 1993) were followed. The parents were planted at five-day intervals to synchronise flowering. Based on the synchronicity of blooming between the parents, hybridization was initiated at the start of flowering bet ween the parents. The operation was conducted in the afternoon (3 pm to 6.30 pm). For emasculation, only those flower buds that were expected to blossom the next morning were chosen. Emasculated flower buds were covered with bag and labelled correctly (**Fig.3**). The next morning, emasculated flowers were pollinated between 4:30 and 7:30 a.m. After pollination, the flowers were again covered with bag and labelled with precision. After three days of cross-pollination, the bags were removed to allow the capsules to develop properly. Then, each capsule that had been cross-fertilized was tagged for identification reasons. These mature capsules were collected with hybrid seeds.

Statistical methods: The combining ability analysis was done as per Griffing's Method 2 Model 1 (Griffing, 1956). Heterosis expressed as percent increase or decrease in hybrid (F_1) over its mid parent value and better parent value in the desirable direction was estimated for various traits as per the formula Relative Heterosis = $100 \times [(F1-MP) / MP]$ suggested by Briggle 1963, Better Parent Heterosis = 100 × [(F1-BP) / BP] suggested by Fonseca and Patterson 1968 Where F_1 = mean hybrid performance, BP = mean performance of better parents and MP = mean performance of mid parent.

The t-test was applied to determine significant difference of F_1 hybrid mean from respective mid parent and better parent values using formulae proposed by Wynne *et al.*1970. The data were analysed in the computer using the Windostat version 8.6 from Indostat service Hyderabad, India. Components of variance due to GCA and SCA were estimated from the expectation of mean squares of the ANOVA for combining ability. The estimates were used to compute predictability factors following Baker (1978).

Predictability Factor (PF) = $2V_{GCA}/(2V_{GCA}+V_{SCA})$

The predictability factor indicates the relative importance of additive gene action in predicting the expression of characters in the progenies.

The data were analysed in the computer using Statistical Package for Agricultural Research (SPAR-I) developed at Indian Statistical Research Institute, New Delhi and also the Windostat version 8.6 from Indostat service Hyderabad, India.

Table 2. Combined analysis of variance for diallel crosses for some quantitative characters

 $*, **$, $***$: Significant at $p = 0.05, 0.01$, and 0.001 respectively

RESULTS AND DISCUSSION

The analysis of variance (**Table 2**) revealed highly significant difference among the parents for all morphological traits with the exception of secondary branches and siliqua length (cm), indicating wide diversity among the parental material used in the present study. The effects of GCA and SCA on all morphological features were statistically significant (Singh *et al.,* 2022), as shown in **Table 3**. This demonstrated the significance of additive and non-additive gene activity in the transmission of all the morphological characteristics. The F_{1s} varied in all characteristics except primary and secondary branches, siliqua length, and 1000 seed weight. The performances of the number of siliqua in each plant, the length of the beak, the number of days to maturity, and the weight of 1000 seeds were significantly different between the parents and the hybrids (Lakshman *et al.,* 2019, Sharma *et al.,* 2020). Hence, genetic variability studies are vital for parent selection in hybridization (Singh *et al.,* 2016a, b). Once genetic variability is known, crop improvement is attainable using an appropriate selection approach. Increasing total yield is simpler by selecting components for yield. According to variation analysis, all evaluated characteristics had a significant genotype influence. This shows that parents and hybrids have enough genetic variation for a full combining ability investigation.

In the F1 generation, ANOVA for combining ability (**Table 3**) demonstrated that variance related to GCA and SCA were significant for all traits except numbers of seeds per siliqua at GCA. This demonstrated the importance of both additive and non-additive gene action in character inheritance. The variance due to SCA was found to be considerably greater than that of GCA for all characters indicating the greater influence of non-additive gene action for exploitation of heterosis (**Table 3**). The results are in agreement with the studies of Chaudhary *et al.* 2019. Most features have a lower ratio, indicating the prevalence of non-additive genes effects were more important than additive effects. Tiwari 2019; Meena *et al.* 2022; Ahmad *et al.* 2022 and Khan *et al.* 2023 also emphasized higher portion of non-additive gene effects in genetic control of seed yield in mustard and believed that selection for improving this trait must be delayed until

later breeding generations. The predictability ratios shown in **Table 3** revealed that, out of the 12 morphological characteristics, the number of secondary branches was predominantly controlled by additive type of gene action. Findings of Kumar and Pandit 2022, indicated a higher contribution of the additive component for number of primary and secondary branches per plant. In accordance with previous studies (Lakshman *et al.,* 2019;Singh *et al.,* 2019; So *et al.,* 2022), additive and non-additive gene effects were reported in mustard. Taking into account all variance estimates, it can be concluded that the number of secondary branches plant¹ and the number of siliqua on the main shoot were controlled primarily by additive gene action, and transgressive breeding may be useful for this trait, whereas seed yield plant⁻¹, siliqua plant⁻¹, and days to maturity were controlled primarily by non-additive gene action, and heterosis breeding is the preferred method for these traits.

The GCA-effects and *per se* performance of the seven parents are shown in **Table 4**. PM-30 was found to be good general combiner for plant height, primary branches, number of siliqua on the main shoot, and seed yield per plant. Other parent, Gujarat Mustard-3 was also good general combiner for number of siliqua per plant and seed yield per plant. Pusa Mahak exhibited good general combiner for primary branches, no. of siliqua per plant, beak length, and seed yield per plant. Considering *per se* performance of these cultivars and their GCA effect on grain yield, selection from progenies of crosses involving the above cultivars will not only improve grain yield, but also increase genetic efficiency of selection. The finding is in line with the report of Chaudhari *et al.* (2022); Singh *et al.* (2022) and Kaur *et al.* (2022). The success of every plant breeding endeavor rests on the selection of suitable parents for hybridization. The GCA is often correlated with genes and modifiable variables (Sprague and Tatum 1942, Shah *et al.,* 2021). These parents may also be used for hybridization or repeated crossing to generate high-yield hybrid varieties or background selection of transgressive segregants to generate pure-line mustard types. Early-generation selection has the potential to enhance additive gene effects for grain production and the majority of yield component characteristics. Based

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Table 5. Scoring of parents in respect of rank in GCA effects for quantitative characters (F₁)

on their GCA effect, each parent was assigned a score (**Table 5**) for each characteristic. "+1" was awarded for any significant GCA effects in the desired direction. The value '-1' was assigned to negative GCA impacts. The score for insignificant GCA effects was 0. None of the seven parents had a favourable GCA for every morphological trait. According to GCA, Pusa Mahak was the best combiner in general with positive score (+3). Genotype Rohini was the poorest general combiner with maximum negative score (-5). Negative GCA score were also observed in Varuna and Jawahar mustard.

In general, the vast majority of crosses in **Table 6** with substantial SCA-effects for any morphological trait also exhibited excellent *per se* performance in the F₁ generation. In the F_1 generation, Jawahar mustard × PM-30 (2.874), Gujarat Mustard-3× Rohini (1.519),Varuna× Rohini, Kranti× PM-30, Kranti× Pusa Mahak, Gujarat Mustard-3 × Jawahar Mustard, and Jawahar Mustard × Pusa Mahak exhibited high *per se* performance and significant SCA-effects for seed yield per plant.These crosses also exhibited higher yield and one of the parents in each cross was a good general combiner indicating that such combinations are expected to produce desirable transgressive segregants. Results obtained from this study agree with results obtained by Singh *et al*. 2022; Ahmad *et al.* 2022 and Devi and Dutta,2020. The cross Varuna× Rohini demonstrated a highly significant SCA effect for plant height, while Varuna× Kranti, Kranti× PM-30, Kranti× Pusa Mahak, Gujarat Mustard-3 × Jawahar Mustard, Jawahar Mustard × PM-30, and Jawahar Mustard × Pusa Mahak demonstrated both high *per se* performance and positive and significant SCA-effects for plant height. Kranti× PM-30 demonstrated outstanding performance *per se* and positive and statistically significant SCAeffects for primary branches. Gujarat mustard-3 × Pusa Mahak exhibited excellent *per se* performance as well as favourable and substantial SCA-effects on secondary branches. For days to 50 percent flowering, Jawahar Mustard × Pusa Mahak demonstrated a high *per se*

performance and a favourable and significant SCA-effect. Kranti× Pusa Mahak had the highest per se performance and the most favourable and considerable SCA-effect for the siliqua on the main shoot, followed by Jawahar Mustard × PM-30 and Gujarat Mustard-3 × Rohini. The number of siliqua per plants in the cross Jawahar Mustard × PM-30 exhibited high per se performance and favourable and considerable SCA-effects, followed by the crosses Varuna× Jawahar Mustard, Gujarat Mustard-3 × Pusa Mahak, and Kranti× PM-30. Varuna× Rohini demonstrated best performance and a statistically significant, positive SCA-effect for beak length, followed by Kranti× Jawahar Mustard. Jawahar Mustard × Pusa Mahak was the most significant in terms of SCA influence for days to maturity, followed by Varuna× Gujarat Mustard-3 and Rohini × PusaMahak. Only one cross between Kranti and Gujarat Mustard-3 was significant for 1000 seed weight. Higher SCA effects observed in this cross where one of the parent had average or good GCA, suggested that additive x dominant gene interaction was involved in governing this trait. The significant negative value for estimates of SCA effects for seed yield per plant was shown by Kranti × Gujarat mustard-3 (-1.524), Gujarat mustard-3× PM-30 (-1.137), Rohini × PM-30 (-2.110), Rohini × PusaMahak (-1.268), PM-30× PusaMahak (-1.389).Overall mean and range of the parents, and F_1 s as well as the difference between the F_1 mean and parental mean, are presented in Fig 1.SCA effects, which are considered to represent non-additive components of genetic variation, are useful for discerning the genetic value of crosses. Several crosses had substantial and acceptable SCA effects for one or more components, but none was an effective combiner for all F_1 characteristics. The crosses Kranti \times Pusa Mahak, Jawahar Mustard × PM-30 and Kranti × PM-30 were promising for seed yield per plant. The F_1 mean was greater than the parental mean for the number of siliqua on the main shoot, medium to low for days to maturity and days to 50 percent flowering, and very low for plant height, secondary branches, seed yield per plant, and primary branches.

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*, *** : Significant at $p = 0.05$, 0.01, and 0.001 respectively

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Table 6. (Continued)

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*, ***, $\ddot{\hspace{2mm}}$: Significant at p = 0.05, 0.01,and 0.001 respectively

*, **, ***: Significant at p = 0.05, 0.01, and 0.001 respectively

Table 7. Heterosis over mid and better parent for 12 quantitative characters in the F, generation **Table 7. Heterosis over mid and better parent for 12 quantitative characters in the F1 generation**

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*, **, *** : Significant at p = 0.05, 0.01,and 0.001 respectively. Figures in the parenthesis indicate significant values**(Contd.)**

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*, **, ***: Significant at p = 0.05, 0.01,and 0.001 respectively. Figures in the parenthesis indicate significant value

Heterosis: The magnitude of heterosis was estimated for all the 12 morphological traits and the same is furnished in **Table 7**. Heterosis over mid-parent for plant height ranged from -14.05 (Rohini × PM-30) to 15.09 percentage (Gujarat mustard-3 × Jawahar mustard) whereas heterosis over better-parent ranged from -14.65 (Rohini × PM-30) to 10.34 percentage (Gujarat mustard-3 × Jawahar mustard). Among the 21 F_{1} s, eight showed positive and significant heterosis over mid-parental and also eight hybrids exhibited positive and significant heterosis over the better-parental value. One cross exhibited negative and significant mid parent heterosis while four crosses showed negative and significant better-parent heterosis for this trait. Crosses with significant and desirable betterparent heterosis along with their specific combining ability effects for different characters, were computed to identify the superior cross combinations for their potential use in hybrid breeding. This experiment showed the presence of significant desirable better-parent heterosis for a good number of crosses for different characters. Singh *et al.* 2022 reported a heterobeltiosis of 51.84. The differences between the estimated heterosis values in this study and those reported previously might be due to the use of different parental combinations.Depending on breeding objectives, both positive and negative heterosis might be advantageous. Positive heterosis is often desired for yield (Singh *et al.,* 2022; Sunny *et al.,* 2022), whereas negative heterosis is desired for early flowering and short plant height (Lamkey and Edwards 1999). Negative heterosis for plant height reduces the likelihood of hybrids lodging, but positive heterosis, although increasing the risk of hybrids lodging, may increase yield, as shown by the positive correlation between plant height and seed production. Numerous studies (Lakshman *et al.,* 2018;Singh *et al.,* 2022) have found negative

heterosis in the height of mustard plants. Grafius, 1959 argues that heterosis in grain production is the result of contemporaneous increase in its many components.

In addition to morphological yield components, other variables may influence mustard seed heterosis. **Table 6** compares better-parent heterosis and SCAeffects. A Majority of the crosses with substantial heterosis in the desired direction displayed substantial SCA effects. It demonstrated the non-additive gene activity's function in heterosis. High GCA (strong GCAeffect in intended direction) or low GCA were assigned to parents (non-significant and significant GCA effects in the undesired direction). Maximum number of hybrids with significant heterosis involved High × Low *gca*parents (55.17%) followed by High × High *gca*-parents (41.38%) and Low × Low *gca*- parents (3.45%) in **Table 8**.The research indicated that parental variability in GCA-effects had a major role in heterosis. Crosses involving at least one parent with a high GCA-effect can only produce exceptional segregants if the additive genetic system in the excellent general combiner and the epistatic effects in the other parent cooperate to maximise the desired plant feature (Singh and Choudary 1995).

Estimates of mustard's combining capacity indicate a substantial opportunity to increase yield and contributing attributes. Experiments on heterosis revealed that all genotypes under investigation had genetic variationthat might be used for both direct selection and hybridization followed by selection.

The study indicated that the nature of gene action indicated that, with the exception of the number of secondary branches and the number of siliqua on the

main shoot, the majority of the other features were governed by a non-additive form of gene action, for which heterosis breeding would be most efficient. The pedigree approach could be used to improve secondary branches per plant, since this attribute was mostly under additive gene regulation. According to the GCA analysis, Gujarat Mustard-3, PM-30, and Pusa Mahak were the best overall general combiners for all morphological features. In terms of seed yield per plant, the analysis of heterosis indicated the three best F1 hybrids to be Jawahar Mustard × PM-30, Varuna×Jawahar Mustard, Gujarath Mustard-3 × PM-30, all of which exhibited high significant heterosis over the mid-parent. These hybrids may be evaluated at many sites, produced as commercial hybrids, or progressed for selfing to isolate transgressive segregants or homozygous lines for use in breeding programmes.

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