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# Efficiency of Molecular Diversity vs Phenotypic Diversity in Attaining Heterosis -A Case Study with Tomato

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***Data Availability Statement:*** Legal restrictions are imposed on the public sharing of raw data. However, authors have full right to transfer or share the data in raw form upon request subject to either meeting the conditions of the original consents and the original research study. Further, access of data needs to meet whether the user complies with the ethical and legal obligations as data controllers to allow for secondary use of the data outside of the original study.

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## Abstract

Presently, both the traditional (phenotypic diversity) and modern tools (molecular markers) are being used to select diverse parents in crop improvement programmes. Several reports are available detailing parallelism between these two types of diversity. However, reports on comparative efficiency of these two types of diversity in terms of attainment of heterosis are lacking. In this context, present investigation was carried out to compare the efficiency of phenotypic diversity vs molecular diversity in realizing genetic components (general combining ability effect, specific combining ability effect and heterosis) determining hybrid performances in tomato. Sixty six tomato entries were subjected to phenotypic- and molecular-diversity assessment using nine phenotypic traits and 29 SSR markers, respectively. Accordingly, two sets of dendrograms were developed. One genotype from each cluster of two dendrograms was selected separately, crossed in half diallel fashion and evaluated in three diverse regions of India. 'Desirability quotient' was calculated for general combining ability effect among two set of parents, and for specific combining ability effect and commercial heterosis among two sets of crosses for important yield traits. Desirability quotient was calculated based on mean desirable genetic parameters (GCA effect, SCA effect and commercial heterosis) and proportion of parents depicting desirable genetic parameters. Molecular diversity-based crosses possessed higher desirability quotient of specific combining ability effect and desirability quotient of heterosis for yield traits under study. The study revealed efficiency of molecular markers in comparison to phenotypic traits in identifying diverse groups possessing higher non-additive gene action and ultimately leading to higher heterosis in tomato.

***Keywords:*** Desirability quotient, diversity, fruit yield, heterosis, SSR

## 1. Introduction

Genetic diversity is the base of all crop improvement programmes. Genetic diversity is related to manifestation of heterosis (Huyen, 2016), an important genetic phenomenon, which results into yield improvement, quality enhancement, elevated tolerance towards biotic and abiotic-stresses, and increased resilience to changing climatic conditions (Bhandari et al., 2017). The diverse lines are of special importance in plant breeding particularly in context of unpredictable climate change (FAO,

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2015) as a source of favourable gene complexes conferring buffering capacity towards new threats. The genetic diversity or 'within-species' diversity confers the survival and adaptability (Rao and Hodgkin, 2002) in variable environments including different locations or different years in a particular location. The major strides in improvement of different crops have been possible as a result of utilization of genetic diversity existing in different crops. However, the commercial varieties of different crops released so far, especially in Indian context, have considerably narrow genetic base resulting into plateaued yield. This necessitates collection and evaluation of new germplasms and their judicious utilization in crop improvement programmes.

Conventionally, genetic diversity is assessed using phenotypic traits and quantitatively assessed using multivariate techniques like $D^2$ analysis and Principal Component Analysis. Genetic diversity assessment using phenotypic evaluation is easy and cheap as it does not require sophisticated tools and preparatory techniques (Sinha and Kumaravadivel, 2015). However, phenotypic measure of genetic diversity suffers from the limitation of few morphological markers and being influenced by environmental factors. In recent years, DNA markers are being increasingly used for assessment of diversity existing among genotypes at molecular level (Fu, 2015). These are considered more efficient on account of being neutral to environment influences (Ferreira, 2006). However DNA marker-based diversity analysis suffers from the limitation of being dependent on only one feature *i.e.* genomic sequences unlike phenotypic evaluation, which is based on several morphological and developmental features of the plant.

Both the methods of diversity analysis have their own benefits and limitations. Many reports are available detailing coherence of these two types of diversity in different crops. However, their comparative efficiency in realizing the combining abilities and heterosis for important economic traits are lacking. Sufficient reports are available indicating association between heterosis and phenotypic (Pandey et al., 2015) or molecular diversity individually. Simultaneous comparison of association between heterosis vs phenotypic diversity/molecular diversity is nowhere reported. Hence, the present investigation is an attempt to compare phenotypic- and molecular diversity in relation to gene effects and heterosis. This type of study can be helpful for plant breeder in deciding the diversity analysis tool to be used for achieving better performances in resultant hybrids.

## 2. Materials and Methods

The test material consisted of 66 genotypes of tomato including Indian and exotic lines. The tomato germplasm lines were evaluated in *rabi*, 2014 at Vegetable Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi (India). The experimental site is located in the middle Gangetic plains in the Eastern part of the state of Uttar Pradesh at 25°19" 59" N latitude, 83°00' 00" E longitude and at elevation of 77 m above mean sea level. Phenotypic diversity was assessed using $D^2$ analysis (Mahalanobis, 1936) based on nine important economic traits viz., days to 50% flowering, plant height, number of primary branches plant$^{-1}$, number of fruits cluster$^{-1}$, number of fruits plant$^{-1}$, average fruit weight, number of locules fruit$^{-1}$, number of seeds fruit$^{-1}$ and fruit yield plant$^{-1}$. Molecular diversity among germplasm lines was assessed using 29 SSR primers covering all the 12 chromosomes. Clustering was done using the symmetric matrix of similarity coefficient and cluster obtained based on unweighted pair group with arithmetic mean (UPGMA) using SHAN module of NTSYS software version 2.0 (Rohlf, 1998).

Based on these studies, two sets of dendrograms were developed. The tomato germplasm (66) were clustered into five and seven groups based on $D^2$ analysis and molecular diversity analysis, respectively. One parent from each cluster from two dendrograms was selected separately and two sets of diallel crosses excluding reciprocals were attempted. The resultant progenies were termed as 1st set for phenotypic diversity-based progenies and as 2nd set for molecular diversity-based progenies. Both the sets of hybrids along with respective parents and a commercial hybrid check To 3150, (A Syngenta Private Ltd. hybrid.) were evaluated at three locations (Pratapgarh, Varanasi and Burdwan) for a period of two years (2015-16 and 2016-17). The locations Pratapgarh and Varanasi represented middle Gangetic plain zone, and the Burdwan location represented lower Gangetic plain zone of India. The data over two years per location were pooled and analysed.

The combining ability analysis was carried out by the procedure given by Griffing (1956). Commercial heterosis was estimated using the formulae as per Kempthorne (1957):

Commercial heterosis=$((\overline{F}_1-\overline{CC})/(\overline{CC})\times100$

Where $\overline{F}_1$=Mean performance of $F_1$ for a trait, $\overline{CC}$=Mean performance of hybrid or commercial check.

A relative term 'Desirability Quotient'is being introduced in this study which can be used for comparing the two sets of parents/crosses for GCA/SCA effect and commercial heterosis. The Desirability quotient was calculated as below:

DQ=$\mu\times p\times100$

Where μ=Mean of significant GCA effect, SCA effect or heterosis (in desirable direction), and p=Proportion of parents/crosses revealing significance (in desirable direction).

## 3. Results and Discussion

The diversity analysis using $D^2$ statistic and SSR markers grouped 66 genotypes into five and seven clusters, respectively. Accordingly, five and seven parents were selected from two dendrograms representing different clusters and half-diallel crosses were made. Desirability quotient of GCA effect, SCA effect and commercial heterosis for two sets of parents was calculated for six important yield traits viz., plant height, number of primary branches plant$^{-1}$, number of fruits plant$^{-1}$, average fruit weight, number of seeds fruit$^{-1}$ and fruit yield plant$^{-1}$. The results are presented in following paragraphs:

### *3.1. General combining ability effect*

The 1st set of parents (phenotypically diverse parents) recorded higher desirability quotient for all six traits *viz.*, plant height, number of primary branches plant$^{-1}$, number of fruits plant$^{-1}$, number of seeds fruit$^{-1}$ and fruit yield plant$^{-1}$in all the locations (Table 1). Present investigation revealed that phenotypically diverse parents possessed higher desirability quotient of GCA effects than that of molecular diversity-based parents for all the traits in all locations. This indicated that the parents selected based on phenotypic diversity possessed higher proportion of additive gene action while, molecular diversity-based parents possessed higher proportion of non-additive gene action.

Table 1: Desirability quotient of gca effects for two sets of parents for different yield traits

| Trait | Plant height | | | | | | No. of primary branches plant$^{-1}$ | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Location | Pratapgarh | | Varanasi | | Burdwan | | Pratapgarh | | Varanasi | | Burdwan | |
| Parameter | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set |
| Mean (desirable) | 16.90 | 3.93 | 15.87 | 5.63 | 17.40 | 2.63 | 1.04 | 0.20 | 1.10 | 0.25 | 0.90 | 0.25 |
| Proportion of desirable parents | 1.00 | 0.29 | 0.20 | 0.43 | 0.20 | 0.14 | 0.20 | 0.43 | 0.20 | 0.57 | 0.20 | 0.57 |
| Desirability Quotient | 1690.0 | 114.0 | 317.4 | 242.1 | 348.0 | 36.8 | 20.8 | 8.6 | 22.0 | 14.3 | 18.0 | 14.3 |
| | No. of fruits plant$^{-1}$ | | | | | | Average fruit weight | | | | | |
| Mean (desirable) | 20.60 | 2.07 | 18.10 | 2.43 | 19.40 | 1.70 | 4.92 | 6.10 | 6.84 | 4.37 | 5.70 | 5.10 |
| Proportion of desirable crosses | 0.20 | 0.29 | 0.20 | 0.14 | 0.20 | 0.57 | 0.80 | 0.29 | 0.60 | 0.43 | 0.80 | 0.29 |
| Desirability Quotient | 412.0 | 60.0 | 362.0 | 34.0 | 388.0 | 96.9 | 393.6 | 176.9 | 410.4 | 187.9 | 456.0 | 147.9 |
| | No. of seeds fruit$^{-1}$ | | | | | | Fruit yield plant$^{-1}$ | | | | | |
| Mean (desirable) | 12.50 | 7.30 | 17.20 | 5.60 | 9.60 | 8.70 | 0.20 | 0.20 | 0.20 | 0.10 | 0.2 | 0.2 |
| Proportion of desirable crosses | 0.60 | 0.29 | 0.40 | 0.29 | 0.80 | 0.29 | 0.80 | 0.29 | 0.80 | 0.57 | 0.80 | 0.14 |
| Desirability Quotient | 750.0 | 211.7 | 688.0 | 162.4 | 768.0 | 252.3 | 16.0 | 5.8 | 16.0 | 5.7 | 16.0 | 2.8 |

### *3.2. Specific combining ability effect*

The 2nd set of hybrids exhibited higher desirability quotient of SCA effect than that of 1st set of hybrids for plant height in all the locations. For the trait, number of primary branches plant$^{-1}$, higher desirability quotient of SCA effect was noted by 2nd set of crosses at Pratapgarh only (Table 2). These behavioural differences may be ascribed to differential expression of genotypes in different location. For the trait number of fruits plant$^{-1}$, 1st set of hybrids recorded higher desirability quotient in all the locations. The 2nd set of hybrids recorded higher desirability quotient of SCA effect for average fruit weight (at Varanasi and Burdwan) and number of seeds fruit$^{-1}$ (at Pratapgarh). For the trait fruit yield plant$^{-1}$, 2nd set of hybrids recorded higher desirability quotient for SCA effect in all the locations. The molecular diversity-based hybrids recorded higher desirability quotient of SCA effect for plant height and fruit yield plant$^{-1}$ universally. In contrast, phenotypic diversity based hybrids recorded higher desirability quotient of SCA effect for number of fruits plant$^{-1}$ in all the locations.

### *3.3. Heterosis*

The 2nd set of hybrids revealed higher desirability quotient than that of 1st set of hybrids for plant height at Varanasi location only (Table 3). The 1st set of hybrids revealed higher desirability quotient for number of primary branches plant$^{-1}$ and number of fruits plant$^{-1}$ in all the locations. In contrast, 2nd set of hybrids revealed universally higher desirability quotient of heterosis for economically important traits like number of seeds fruit$^{-1}$, average fruit weight and fruit yield plant$^{-1}$. For the traits average fruit weight and fruit yield plant$^{-1}$, none of the crosses among 1st set revealed desirable heterosis in any location when compared to the commercial check. As a result, 1st set of crosses recorded null value of desirability quotient for these traits. Consequently, 2nd set of hybrids consistently recorded higher desirability quotient than that of 1st set of hybrids for these two traits.

Higher desirability quotient of heterosis for major economic traits like number of seeds fruit$^{-1}$, average fruit weight and fruit yield plant$^{-1}$ was noted by crosses derived from crossing molecular diverse parents. This may be attributed to the specific heterozygosity of positive markers (Pheirim, 2017). The 1st set of hybrids exhibited higher desirability quotient of heterosis for plant height, number of primary branches

Table 2: Desirability quotient of sca effects for two sets of crosses for different yield traits

| Trait | Plant height | | | | | | No. of primary branches plant$^{-1}$ | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Location | Pratapgarh | | Varanasi | | Burdwan | | Pratapgarh | | Varanasi | | Burdwan | |
| Parameter | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set |
| Mean (desirable) | 6.39 | 13.71 | 11.70 | 14.81 | 16.38 | 16.51 | 0.47 | 0.41 | 0.50 | 0.48 | 0.52 | 0.44 |
| Proportion of desirable parents | 0.20 | 0.33 | 0.30 | 0.62 | 0.40 | 0.43 | 0.30 | 42.9 | 0.60 | 38.1 | 0.40 | 33.3 |
| Desirability quotient | 127.8 | 452.4 | 351.0 | 918.2 | 655.2 | 709.9 | 14.1 | 17.7 | 30.0 | 18.4 | 20.8 | 14.8 |
| | No. of fruits plant$^{-1}$ | | | | | | Average fruit weight | | | | | |
| Mean (desirable) | 10.5 | 6.39 | 9.09 | 5.01 | 6.43 | 6.92 | 4.64 | 4.76 | 3.62 | 8.15 | 4.70 | 5.04 |
| Proportion of desirable crosses | 0.40 | 0.43 | 0.40 | 0.38 | 0.60 | 0.48 | 0.30 | 0.29 | 0.40 | 0.29 | 0.30 | 0.52 |
| Desirability quotient | 420.0 | 274.8 | 363.6 | 190.4 | 385.8 | 332.2 | 139.2 | 138.0 | 144.8 | 236.4 | 141.0 | 262.1 |
| | No. of seeds fruit$^{-1}$ | | | | | | Fruit yield plant$^{-1}$ | | | | | |
| Mean (desirable) | 18.59 | 18.5 | 22.66 | 15.7 | 17.49 | 14.99 | 0.21 | 0.30 | 0.21 | 0.31 | 0.38 | 0.29 |
| Proportion of desirable crosses | 0.20 | 0.29 | 0.30 | 0.38 | 0.30 | 0.33 | 0.30 | 0.52 | 0.40 | 0.38 | 0.40 | 0.67 |
| Desirability quotient | 371.8 | 536.5 | 679.8 | 596.6 | 524.7 | 494.7 | 6.3 | 15.6 | 8.4 | 11.8 | 15.2 | 19.4 |

Table 3: Desirability quotient of commercial heterosis for two sets of crosses for different yield traits

| Trait | Plant height | | | | | | No. of primary branches plant$^{-1}$ | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Location | Pratapgarh | | Varanasi | | Burdwan | | Pratapgarh | | Varanasi | | Burdwan | |
| Parameter | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set | 1st set | 2nd set |
| Mean (desirable) | 13.86 | 16.82 | 16.38 | 23.53 | 27.06 | 22.42 | 28.6 | 11.5 | 24.5 | 14.6 | 18.6 | 12.2 |
| Proportion of desirable parents | 0.30 | 0.05 | 0.20 | 0.19 | 0.80 | 0.43 | 0.40 | 0.24 | 0.70 | 0.57 | 0.60 | 0.10 |
| Desirability quotient | 415.8 | 84.1 | 327.6 | 447.1 | 2164.8 | 964.1 | 1144.0 | 276.0 | 1715.0 | 832.2 | 1116.0 | 122.0 |
| | No. of fruits plant$^{-1}$ | | | | | | Average fruit weight | | | | | |
| Mean (desirable) | 53.6 | 14.4 | 38.2 | 12.46 | 45.1 | 20.85 | - | 15.3 | - | 18.76 | - | 9.87 |
| Proportion of desirable crosses | 0.50 | 0.40 | 0.50 | 0.05 | 0.90 | 0.57 | - | 0.40 | - | 0.05 | - | 0.05 |
| Desirability quotient | 2680.0 | 576.0 | 1910.0 | 62.3 | 4059.0 | 1188.5 | - | 612.0 | - | 93.8 | - | 49.35 |
| | No. of seeds fruit$^{-1}$ | | | | | | Fruit yield plant$^{-1}$ | | | | | |
| Mean (desirable) | 22.53 | 33.8 | 26.6 | 28.02 | 13.75 | 32.4 | - | 11.7 | - | 5.94 | - | 8.12 |
| Proportion of desirable crosses | 0.20 | 0.62 | 0.30 | 0.57 | 0.20 | 0.43 | - | 33.3 | - | 0.05 | - | 0.10 |
| Desirability quotient | 450.6 | 2095.6 | 798.0 | 1597.1 | 275.0 | 1393.2 | - | 388.6 | - | 29.7 | - | 81.2 |

plant$^{-1}$ and number of fruits plant$^{-1}$. The 2nd set of hybrids consistently recorded higher desirability quotient of heterosis for important yield traits namely average fruit weight, number of seeds fruit$^{-1}$ and fruit yield plant$^{-1}$. The higher desirability quotient of heterosis for 1st set of hybrids for number of fruits plant$^{-1}$ in all locations may be attributed to the higher desirability quotient of SCA effect for the trait. The 1st set of hybrids failed to record higher desirability quotient of heterosis for fruit yield plant$^{-1}$ despite higher desirability quotient for number of fruits plant$^{-1}$ indicating non-coherence between these traits. The better score of desirability quotient of commercial heterosis for number of seeds fruit$^{-1}$ and fruit yield plant$^{-1}$ may be linked to better score of average fruit weight. The study indicates that average fruit weight might have more contribution towards fruit yield.

The present investigation revealed higher efficacy of molecular markers in identifying diverse groups possessing higher non-additive gene action and ultimately the higher heterosis for important yield traits in tomato. This is in accordance to the reports of Pandey et al. (2015) who established the necessity of molecular markers in determination of genetic diversity in context of non-consistent correlation between phenotypic diversity and heterosis. Similarly, Sud et al. (2010) indicated strong association of yield heterosis with SSR-based genetic diversity. In contrast, Cebolla-Cornejo et al. (2013) emphasized the need of morpho-agronomical and quality characterization over molecular characterization in the *ex-situ* management of genetic resources in Spanish tomato landraces. Ezekiel et al. (2011) reported higher efficiency of molecular markers over phenotypic markers in determination of varietal identity. The present study establishes that fruit weight has greater association with fruit yield in tomato and is supported by reports of de Souza et al. (2012). In addition, the study indicated higher efficacy of molecular markers in identifying genotypes to be used in hybrid breeding programme.

## 4. Conclusion

The present study indicated higher efficiency of molecular diversity than that of phenotypic markers in attaining heterosis for yield traits in tomato.

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