

Genetic Mechanisms Underlying Heterotic Response and Inbreeding Decline for Yield and Grain Biofortification Traits in Rice (*Oryza sativa* L.)

Usharani G¹, Neeraja C N², Nagesh P³, Ramya R⁴, Hemanth K R M⁵, Madhubabu P², Rao D S², Babu V R², Bharathi M⁶, Rani CH V D⁷

¹Agricultural Research Station, Karimnagar, PJTSAU, Telangana, India

²Indian Institute of Rice Research, Rajendranagar, Hyderabad, Telangana, India

³Global Maize Programme, CIMMYT, ICRISAT, Hyderabad, Telangana, India

⁴Regional Sugarcane and Rice Research Station, Rudrur, Telangana, India

⁵KLU, Guntur, Andhra Pradesh, India

⁶Department of Genetics and Plant Breeding, College of Agriculture, PJTSAU, Rajendranagar, Hyderabad, Telangana, India

⁷Department of Plant Molecular Biology and Biotechnology, College of Agriculture, PJTSAU, Rajendranagar, Hyderabad, Telangana, India

† Corresponding Author: Usharani G, Email: ushagenetics@gmail.com

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Abstract- The present investigation was undertaken to estimate heterosis, heterobeltiosis, and inbreeding depression for yield, grain quality, and micronutrient traits in the rice cross ‘IR-64 × Ranbir Basmati’. The experimental material comprising parents, F₁, and F₂ generations was evaluated during the kharif season under irrigated conditions in a Randomized Block Design with two replications. Observations were recorded on days to 50 per cent flowering, plant height, productive tillers per plant, panicle length, filled grains per panicle, grain yield per plant, 1000-grain weight, kernel dimensions, length/breadth ratio, iron content, and zinc content. Significant positive heterosis and heterobeltiosis were observed for productive tillers, grain yield per plant, 1000-grain weight, iron content, and zinc content, indicating strong hybrid vigor and superior breeding potential. Grain yield per plant exhibited high positive heterosis along with moderate to high inbreeding depression, suggesting the predominance of non-additive gene action. Grain quality traits recorded comparatively lower inbreeding depression, indicating relatively stable inheritance and additive genetic control. Significant positive heterosis for iron and zinc content demonstrated the possibility of simultaneous improvement of productivity and nutritional quality through hybridization. The results revealed that the cross ‘IR-64 × Ranbir Basmati’ possesses substantial potential for utilization in rice improvement and biofortification breeding programmes.

Keywords: Rice, heterosis, heterobeltiosis, inbreeding depression, grain quality, iron, zinc, biofortification

Introduction

Rice (Rice) is regarded as one of the most indispensable food crops in the world because of its direct contribution to human nutrition and global food security. In many Asian countries, including India, rice serves not only as a staple food but also as an important source of livelihood and rural employment. The crop contributes significantly to caloric intake and daily dietary requirements of millions of people. However, sustaining rice productivity under increasing population pressure and changing environmental conditions remains a major challenge for plant breeders. Simultaneously, there is growing demand for rice varieties

possessing superior grain quality and enhanced nutritional value, particularly with respect to essential micronutrients such as iron and zinc. Micronutrient deficiency has become a serious nutritional concern in developing countries where cereal-based diets dominate daily food consumption. Iron deficiency anemia and zinc malnutrition are among the most prevalent forms of hidden hunger affecting human health worldwide. Deficiency of iron leads to fatigue, reduced immunity, impaired cognitive development, and lower work productivity, whereas zinc deficiency causes growth retardation, poor immune response, and increased vulnerability to diseases. Since polished rice grains generally contain low concentrations of these micronutrients, improving grain nutritional quality through genetic enhancement has become an important objective in rice breeding programmes (Swamy et al., 2021). Biofortification breeding offers a sustainable and cost-effective approach to enhance micronutrient concentration in staple food crops. Unlike dietary supplementation or industrial fortification, biofortified crop varieties provide long-term nutritional benefits without requiring major changes in food habits or recurring economic inputs. Recent advances in rice breeding have demonstrated the existence of sufficient genetic variability for grain iron and zinc content, thereby providing opportunities for the development of nutritionally enriched rice cultivars. Nevertheless, improvement of micronutrient concentration should be achieved without compromising grain yield and grain quality, which remain the primary selection criteria in varietal development programmes. Hybridization is an important breeding approach for combining desirable traits from genetically diverse parents and generating superior recombinants. The success of hybrid breeding largely depends upon the extent of heterosis expressed in the F_1 generation and the nature of gene action controlling the target traits. Heterosis breeding has contributed substantially toward increasing rice productivity through the development of high-yielding hybrids exhibiting superior performance for yield and associated characters. Estimation of heterosis and heterobeltiosis helps identify promising cross combinations capable of producing vigorous hybrids, whereas the study of inbreeding depression provides useful information regarding the relative importance of additive and non-additive gene effects governing trait inheritance (Falconer and Mackay, 1996). The cross 'IR-64 $\times$ Ranbir Basmati' represents a unique genetic combination involving two contrasting parental genotypes. IR-64 is a widely adapted, high-yielding semi-dwarf variety possessing good agronomic performance and stable productivity under diverse environments. In contrast, Ranbir Basmati is valued for its premium grain quality, aroma, and consumer acceptability. Combining these two genetically diverse parents offers opportunities for simultaneous improvement of yield, grain quality, and micronutrient traits. Evaluation of heterotic response in this cross may therefore provide useful insights into the breeding potential of the hybrid and the inheritance pattern of economically important traits. Earlier investigations have reported considerable heterosis for yield and quality traits in rice. Significant positive heterosis for productive tillers, panicle traits, grain yield, and grain weight has been documented by Alam et al. (2004), Pandya and Tripathi (2006), and Borah and Barman (2010). Improvement in grain quality traits through heterosis breeding was also reported by Raju et al. (2003) and Sanjeevkumar et al. (2010). More recently, emphasis has been placed on integrating biofortification traits into mainstream rice breeding programmes to address nutritional insecurity and improve the health benefits of rice. Considering the importance of developing high-yielding and nutritionally enriched rice cultivars, the present investigation was undertaken to study heterosis, heterobeltiosis, and inbreeding depression for yield, grain quality, and micronutrient traits in the rice cross 'IR-64 $\times$ Ranbir Basmati'. The study aimed to identify promising traits and assess the breeding potential of the cross for future rice improvement and biofortification programmes.

Materials and Methods

The present investigation was conducted during the kharif season under irrigated conditions at the Directorate of Rice Research (DRR), ICRISAT campus, Patancheru, Hyderabad, Telangana. The experimental site is located at 17.53°N latitude and 78.27°E longitude with an altitude of approximately 545 m above mean sea level. The region is characterized by semi-arid tropical climatic conditions suitable for rice cultivation under irrigated ecosystems. The experimental material comprised the rice cross 'IR-64 $\times$ Ranbir Basmati'. The parental genotypes were selected based on their contrasting performance for yield, grain quality, and nutritional traits. IR-64 is a high-yielding semi-dwarf cultivar known for its adaptability and stable agronomic performance, whereas Ranbir Basmati is recognized for superior grain quality and aromatic characteristics. Hybridization was carried out using standard clipping and pollination techniques to generate F_1 seeds. The F_1 plants were selfed to obtain the F_2 generation. The parents, F_1 , and F_2 populations were raised in a Randomized Block Design with two replications following recommended agronomic and crop management practices. Observations were recorded on five randomly selected competitive plants in each replication for days to 50 per cent flowering, plant height, panicle length, productive tillers per plant, filled grains per panicle, grain yield per plant, and 1000-grain weight. Grain quality traits including kernel length, kernel breadth, and length/breadth ratio were estimated using a digital seed analyzer. Iron and zinc content in polished rice grains were estimated using Energy Dispersive X-ray Fluorescence Spectrometry (EDXRF) following standard analytical procedures described by Paltridge et al. (2012). Heterosis over mid parent, heterobeltiosis over better parent, and inbreeding depression were estimated using standard biometrical procedures. The significance of heterosis estimates was tested using 't' tests as suggested by Wynne et al. (1970). The magnitude and direction

of heterosis and inbreeding depression were used to understand the nature of gene action governing the inheritance of yield, grain quality, and micronutrient traits.

Results and Discussion

The estimates of heterosis, heterobeltiosis, and inbreeding depression in the cross 'IR-64 × Ranbir Basmati' revealed substantial variability for yield, grain quality, and micronutrient traits, indicating the presence of considerable genetic divergence between the parental genotypes. The expression of heterosis in the F₁ generation and the extent of inbreeding depression in the F₂ generation suggested the involvement of both additive and non-additive gene effects in the inheritance of the studied traits. The results further indicated that the cross possessed significant potential for exploitation in rice improvement and biofortification breeding programmes.

Table 1. Estimates of heterosis, heterobeltiosis and inbreeding depression in the cross 'IR- 64 X Ranbir Basmati'

S.No.	Character	Heterosis percent over		Inbreeding Depression
		MP	BP	
1	Days to 50% flowering	2.13**	3.23**	0.00
2	Plant Height	5.38	35.14**	-5.63
3	Panicle Length	0.61	-5.01**	1.01**
4	Productive Tillers	33.33**	7.69**	12.23**
5	Yield per plant	28.41**	8.21**	28.30**
6	Filled Grains	-4.18**	-15.61**	28.87*
7	1000 Grain Weight	13.26**	7.25**	8.46
8	Kernel Length	3.59**	0.47**	2.69**
9	Kernel Breadth	4.41**	12.55**	2.43**
10	Length/Breadth Ratio	-1.34**	-10.25**	-0.74**
11	Iron	18.72**	13.54**	17.98**
12	Zinc	28.89**	17.91**	12.17**

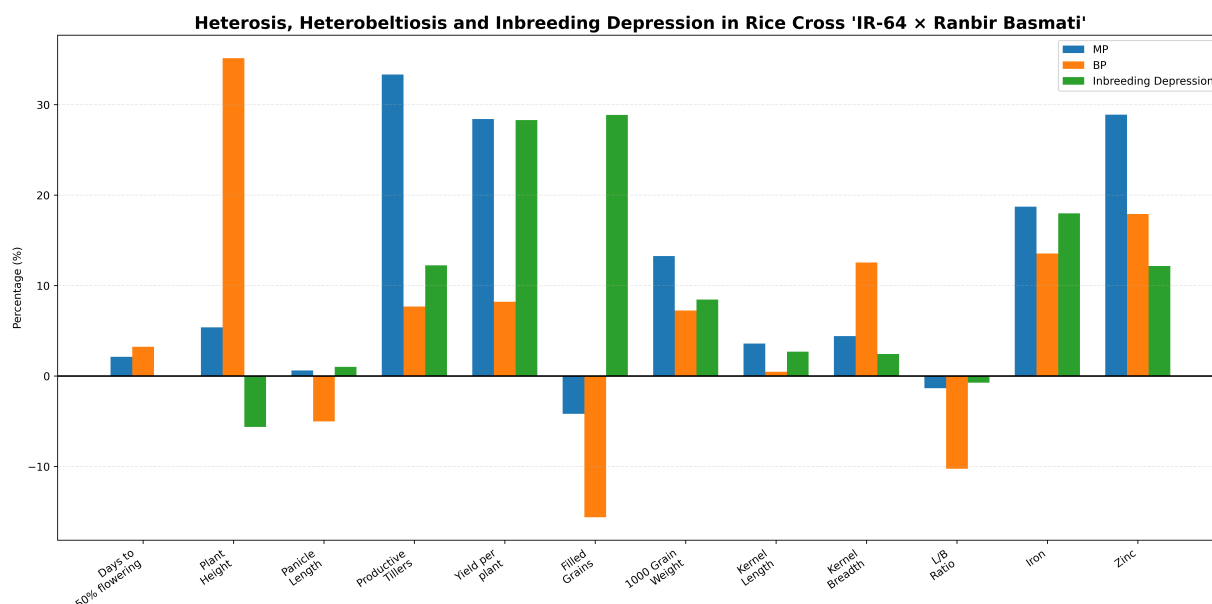


Fig. 1. Comparative estimates of heterosis, heterobeltiosis and inbreeding depression for yield, grain quality and micronutrient traits in the rice cross 'IR-64 × Ranbir Basmati'.

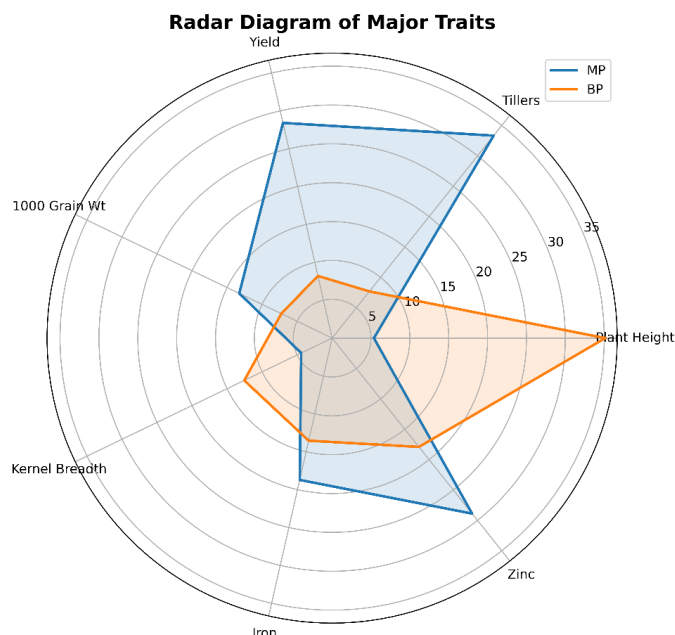


Fig. 2. Radar diagram showing heterotic performance of major yield, grain quality and micronutrient traits in the rice cross 'IR-64 × Ranbir Basmati'

Days to 50 % Flowering

Days to 50 % flowering exhibited significant positive heterosis of 2.13 per cent over the mid parent and 3.23 per cent over the better parent, whereas no appreciable inbreeding depression was observed in the F_2 generation. The positive heterotic response indicated a slight increase in crop duration compared with the parental lines. The absence of substantial inbreeding depression suggested relative stability in the inheritance of flowering behaviour and indicated comparatively lower influence of dominance deviations for this trait. Flowering duration is an important adaptive trait influencing maturity period, cropping intensity, and environmental adaptation. Moderate flowering duration is often preferred in irrigated ecosystems because it allows sufficient vegetative growth and effective grain filling. The results obtained in the present study are in conformity with earlier reports of Pandya and Tripathi (2006), Deoraj et al. (2007), and Saravanan et al. (2006), who also observed variable heterotic responses for flowering duration in rice hybrids. The stable inheritance observed in the present cross indicated that effective selection for flowering duration may be possible in subsequent segregating generations.

Plant Height

Plant height recorded positive heterosis of 5.38 per cent over the mid parent and significant heterobeltiosis of 35.14 per cent over the better parent. Interestingly, negative inbreeding depression (-5.63%) was observed, indicating that the F_2 generation maintained comparable or slightly superior performance relative to the F_1 generation for this trait. The pronounced heterobeltiosis observed for plant height may be attributed to enhanced vegetative vigor resulting from favorable interactions between alleles contributed by the two genetically diverse parents. Plant height is one of the important agronomic traits influencing biomass accumulation, nutrient translocation, and lodging resistance. Although excessive plant height may increase lodging susceptibility, moderate increase in plant stature can contribute toward improved photosynthetic efficiency and dry matter production. The negative inbreeding depression observed in the present study suggested relative genetic stability and possible fixation of favorable allelic combinations controlling plant height. Similar observations were reported earlier by Verma et al. (2004), Saravanan et al. (2006), and Deoraj et al. (2007). The results therefore indicated that the inheritance of plant height in this cross may involve both additive and partial dominance gene effects.

Panicle Length

Panicle length exhibited low positive heterosis of 0.61 per cent over the mid parent but negative heterobeltiosis (-5.01%) over the better parent. The trait showed very low inbreeding depression (1.01%), indicating comparatively stable inheritance and limited breakdown of favorable gene combinations in the F₂ generation. The low magnitude of heterosis suggested that both parental lines possessed nearly similar genetic potential for panicle length. Panicle length is an important yield-contributing trait because longer panicles generally possess greater grain-bearing capacity. The comparatively low inbreeding depression observed in the present investigation indicated predominance of additive genetic effects and greater possibility of improvement through direct selection. Similar findings were reported by Pandya and Tripathi (2006), Borah and Barman (2010), and Gouri Shankar et al. (2010). The stability of this trait across generations suggested that panicle length may be effectively fixed in advanced breeding generations.

Productive Tillers per Plant

Productive tillers per plant recorded substantial positive heterosis of 33.33 per cent over the mid parent and 7.69 per cent over the better parent. The trait also exhibited moderate inbreeding depression (12.23%), indicating the involvement of non-additive gene action. Among the yield-contributing traits studied, productive tillers showed one of the highest magnitudes of heterosis, demonstrating the strong hybrid vigor expressed in the F₁ generation. Productive tillers are directly associated with sink capacity and play a crucial role in determining final grain yield in rice. Increased tillering ability enhances the number of productive panicles per plant and thereby contributes significantly toward overall productivity. The moderate inbreeding depression observed in the F₂ generation suggested partial breakdown of favorable dominant gene interactions responsible for enhanced tillering in the F₁ hybrid. The present findings corroborate the earlier reports of Bharathi (1998), Venkatesan et al. (2008), and Gouri Shankar et al. (2010), who also reported substantial heterosis for productive tillers in rice hybrids. The high heterotic response observed in this cross indicated the usefulness of hybrid breeding approaches for improving tillering ability and grain yield potential.

Filled Grains per Panicle

Filled grains per panicle exhibited negative heterosis of -4.18 per cent over the mid parent and -15.61 per cent over the better parent. However, the trait recorded relatively high inbreeding depression (28.87%), indicating the involvement of dominance and over-dominance effects in its inheritance. The reduction in filled grains per panicle in the F₁ generation suggested that the hybrid combination did not exhibit favorable complementation for this particular trait. Despite the negative heterosis observed for filled grains per panicle, the hybrid recorded superior grain yield due to compensatory improvement in other yield-contributing traits such as productive tillers and 1000-grain weight. Grain yield in rice is governed by complex interactions among several component characters, and therefore improvement in one or more major components may offset the reduction in other traits. The high inbreeding depression observed for filled grains per panicle indicated instability of favorable gene combinations in the segregating generation. Similar heterotic responses for this trait were also reported by Reddy and Nerkar (1995), Sonika et al. (2009), and Panwar and Mashiat Ali (2010). The results suggested that careful selection may be required in later generations to recover desirable recombinants possessing improved grain filling ability.

Grain Yield per Plant

Grain yield per plant exhibited substantial positive heterosis of 28.41 per cent over the mid parent and 8.21 per cent over the better parent, indicating superior hybrid performance and strong expression of hybrid vigor. The trait also recorded high inbreeding depression (28.30%), suggesting the predominance of non-additive gene action such as dominance and over-dominance. Grain yield is a highly complex quantitative trait influenced by several component characters and their interaction with the environment. The superior performance of the F₁ hybrid in the present investigation may largely be attributed to increased productive tillers, enhanced grain weight, and improved vegetative vigor. The marked reduction in performance observed in the F₂ generation indicated partial dissociation of favorable dominant gene combinations during segregation. The present findings are in close agreement with earlier reports of Alam et al. (2004), Ashok et al. (2006), Borah and Barman (2010), and Panwar and Mashiat Ali (2010), who also observed significant positive heterosis for grain yield in rice hybrids. High heterosis coupled with considerable inbreeding depression suggested that exploitation of hybrid vigor through heterosis breeding would be more effective for improving grain yield than direct selection in early segregating generations. The results further demonstrated that the cross 'IR-64 × Ranbir Basmati' possesses substantial breeding potential for developing high-yielding rice hybrids and superior segregants with enhanced productivity.

1000-Grain Weight

The trait 1000-grain weight exhibited significant positive heterosis of 13.26 per cent over the mid parent and 7.25 per cent over the better parent. Inbreeding depression was comparatively low (8.46%), indicating relative stability of gene action controlling grain weight. Positive heterosis for grain weight is particularly desirable because it directly contributes toward increased grain yield and improved market value. The comparatively lower inbreeding depression suggested the involvement of additive and partial dominance effects, thereby indicating better scope for effective selection in segregating generations. The results are in accordance with earlier findings reported by Pandya and Tripathi (2006), Venkatesan et al. (2008), and Gouri Shankar et al. (2010). Improvement in grain weight may also contribute positively toward grain appearance and milling recovery, thereby enhancing commercial acceptability.

Grain Quality Traits

Kernel length exhibited positive heterosis of 3.59 per cent over the mid parent and 0.47 per cent over the better parent, whereas kernel breadth showed positive heterosis over both parental estimates. However, the length/breadth ratio recorded slight negative heterosis. All these quality traits exhibited very low inbreeding depression, indicating comparatively stable inheritance and lower influence of non-additive gene action. Grain quality traits are extremely important in rice because they determine market demand, cooking quality, and consumer preference. Ranbir Basmati, being a premium quality aromatic parent, may have contributed favorable alleles for grain dimensions and grain appearance. The relatively stable inheritance observed for these traits suggested that selection for grain quality may be effectively practiced in early segregating generations. The present findings corroborate the earlier reports of Raju et al. (2003) and Sanjeevkumar et al. (2010), who also reported significant heterotic expression for kernel traits in rice. Improvement of grain quality traits along with high productivity is particularly important for developing commercially acceptable rice varieties.

Iron Content

Iron content recorded significant positive heterosis of 18.72 per cent over the mid parent and 13.54 per cent over the better parent. The trait also exhibited moderate inbreeding depression (17.98%), indicating the involvement of both additive and non-additive gene effects. The significant heterotic response observed for iron concentration suggested the possibility of improving nutritional quality through hybridization between genetically diverse parents. Iron deficiency is one of the major nutritional disorders affecting human populations dependent on cereal-based diets. Therefore, enhancement of grain iron concentration has become an important objective in rice biofortification programmes. The present findings indicated that the cross 'IR-64 × Ranbir Basmati' possesses considerable potential for improving iron content without adversely affecting grain yield. Similar observations were reported by and emphasized the significance of genetic biofortification in combating micronutrient malnutrition. The moderate inbreeding depression observed for iron content further suggested that selection for improved iron concentration may be feasible in advanced generations.

Zinc Content

Among the micronutrient traits studied, zinc content exhibited the highest magnitude of positive heterosis with 28.89 per cent over the mid parent and 17.91 per cent over the better parent. Moderate inbreeding depression (12.17%) indicated partial retention of favorable gene combinations in the F₂ generation. The substantial heterotic response observed for zinc concentration demonstrated the effectiveness of hybridization for improving nutritional quality in rice. Zinc plays an important role in human metabolism, immune function, and growth regulation. Since rice is a staple food for a large proportion of the global population, enhancement of zinc concentration in rice grains can contribute significantly toward reducing hidden hunger. The simultaneous expression of positive heterosis for grain yield and zinc concentration observed in the present study is particularly important because these traits are often negatively associated. The results obtained are in conformity with the findings of who also reported the feasibility of improving grain zinc concentration through breeding approaches. The present investigation therefore demonstrated that the cross 'IR-64 × Ranbir Basmati' possesses substantial potential for the development of nutritionally enriched high-yielding rice cultivars suitable for future biofortification breeding programmes.

Conclusion

The present investigation revealed substantial heterotic variability in the rice cross 'IR-64 × Ranbir Basmati' for yield, grain quality, and micronutrient traits. Significant positive heterosis and heterobeltiosis were observed for productive tillers, grain yield per plant, 1000-grain weight, iron content, and zinc content, indicating superior hybrid performance and considerable breeding potential. Moderate to high inbreeding depression observed for grain yield and productive tillers suggested the predominance of non-additive gene action and highlighted the importance of heterosis breeding for improvement of these traits. In contrast, lower inbreeding depression for grain quality traits indicated relatively stable inheritance and greater scope for effective selection in segregating generations. The simultaneous expression of positive heterosis for grain yield along with iron and zinc content demonstrated the possibility of developing nutritionally enriched rice cultivars through appropriate hybridization and selection strategies. Overall, the cross 'IR-64 × Ranbir Basmati' emerged as a promising genetic combination for the development of high-yielding rice varieties possessing desirable grain quality and enhanced nutritional value, thereby contributing toward sustainable rice production and nutritional security.

REFERENCES

1. Alam MF, Khan MR, Nuruzzaman M, Parvez S, Swaraz AM, Alam I and Ahsan N. 2004. Genetic basis of heterosis and inbreeding depression in rice (*Oryza sativa* L.). *Journal of Zhejiang University Science*. 5(4):406–411.
2. Ashok S, Jyothula DPB and Ratnababu D. 2006. Heterosis and combining ability studies in rice. *Andhra Agricultural Journal*. 53:35–38.
3. Bharathi M. 1998. Heterosis studies in rice hybrids. M.Sc. (Ag.) Thesis, Tamil Nadu Agricultural University, Coimbatore.
4. Borah SP and Barman KK. 2010. Heterosis for yield and its component traits in rice. *Oryza*. 47(1):77–79.
5. Deoraj, Singh NB and Singh AK. 2007. Heterosis and inbreeding depression for yield and quality traits in rice. *Oryza*. 44(1):73–75.
6. Falconer DS and Mackay TFC. 1996. *Introduction to Quantitative Genetics*. 4th Edition. Longman Group Ltd., England.
7. Gouri Shankar VG, Kushwaha UKS and Mishra VK. 2010. Heterosis studies for yield and its components in rice. *Oryza*. 47(1):79–81.
8. Paltridge NG, Palmer LJ, Milham PJ, Guild GE and Stangoulis JCR. 2012. Energy-dispersive X-ray fluorescence analysis of zinc and iron concentration in rice and pearl millet grain. *Plant and Soil*. 361:251–260.
9. Pandya R and Tripathi RS. 2006. Heterosis breeding in rice. *Oryza*. 43(2):87–93.
10. Panwar LL and Mashiat Ali. 2010. Heterosis and inbreeding depression for grain yield and yield components in rice. *Oryza*. 47(3):179–188.
11. Raju CHS, Rao MVB and Reddy TD. 2003. Heterosis for grain quality traits in rice. *Annals of Agricultural Research*. 24(4):1064–1066.
12. Reddy JN and Nerkar YS. 1995. Heterosis studies in rice. *Oryza*. 32:15–18.
13. Sanjeev Kumar B, Singh CM and Suresh BG. 2010. Studies on heterosis for grain quality traits in rice. *Oryza*. 47(4):325–328.
14. Saravanan K, Sabesan T and Thangavel P. 2006. Studies on heterosis in rice. *Plant Archives*. 6(1):287–290.
15. Sonika H, Ramesh B and Kumar V. 2009. Heterosis studies for grain yield and its components in rice. *Crop Research*. 38:145–148.
16. Swamy BPM, Rahman MA, Inabangan-Asilo MA, Amparado A, Manito C, Reinke R and Slamet-Loedin IH. 2021. Advances in breeding for high grain zinc in rice. *Rice*. 14:84. <https://doi.org/10.1186/s12284-021-00530-2>
17. Venkatesan M, Anbuselvam Y and Elangaimannan R. 2008. Heterosis for yield traits in rice. *Crop Improvement*. 35(1):76–79.

18. Verma OP, Srivastava HK and Singh H. 2004. Heterosis and combining ability analysis in rice hybrids. *Indian Journal of Genetics and Plant Breeding*. 64:296–298.
19. Wynne JC, Emery DA and Rice PH. 1970. Combining ability estimates in *Arachis hypogaea* L. II. Field performance of F₁ hybrids. *Crop Science*. 10:713–715.