GENETIC ANALYSIS OF GRAIN IRON AND ZINC CONCENTRATIONS IN SORGHUM (*Sorghum bicolor* L. Moench)

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B.Sc. (Ag.)

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(GENETICS AND PLANT BREEDING)

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BY
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THESIS SUBMITTED TO THE
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CHAIRPERSON: Dr. K. RADHIKA

DEPARTMENT OF GENETICS AND PLANT BREEDING
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CERTIFICATE

Mr. **K. TIRUMALA RAVI KIRAN** has satisfactorily prosecuted the course of research and that thesis entitled **"GENETIC ANALYSIS OF GRAIN IRON AND ZINC CONCENTRATIONS IN SORGHUM (*Sorghum bicolor* L. Moench)"** submitted is the result of original research work and is of sufficiently high standard to warrant its presentation to the examination. I also certify that neither the thesis nor its part thereof has been previously submitted by him for a degree of any University.

Date: **(K. RADHIKA)**

Place: Hyderabad Chairperson

CERTIFICATE

This is to certify that the thesis entitled **"GENETIC ANALYSIS OF GRAIN IRON AND ZINC CONCENTRATIONS IN SORGHUM (*Sorghum bicolor* L. Moench)"** submitted in partial fulfilment of the requirements for the degree of 'Master of Science in Agriculture' of the Acharya N.G. Ranga Agricultural University, Hyderabad is a record of the bonafied original research work carried out by Mr. **K. TIRUMALA RAVI KIRAN** under our guidance and supervision.

No part of the thesis has been submitted by the student for any other degree or diploma. The published part and all assistance received during the course of the investigations have been duly acknowledged by the author of the thesis.

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DECLARATION

I**, K. TIRUMALA RAVI KIRAN** hereby declare that the thesis entitled **"GENETIC ANALYSIS OF GRAIN IRON AND ZINC CONCENTRATIONS IN SORGHUM (*Sorghum bicolor* L. Moench)"** submitted to the **Acharya N.G. Ranga Agricultural University** for the degree of **Master of Science in Agriculture** is the result of original research work done by me. I also declare that no material contained in the thesis has been published earlier in any manner.

Place: Hyderabad **(K. TIRUMALA RAVI KIRAN)**

Date: **I. D. No: RAM/11-31**

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Place: Hyderabad **(K. T. RAVI KIRAN)**

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LIST OF ILLUSTRATIONS

LIST OF SYMBOLS AND ABBREVIATIONS

%	:	Per cent
ANOVA	:	Analysis of Variance
A.P.	:	Andhra Pradesh
B	:	Boron
B_1	:	Progeny of backcross of F_1 with parent 1
B_2	:	Progeny of backcross of F_1 with parent 2
Ca	:	Calcium
C.D.	:	Critical Difference
C.V.	:	Coefficient of Variation
CGIAR	:	Consultative Group on International Agricultural Research
CoV	:	Covariance
Cu	:	Copper
d	:	Additive
EMS	:	Error Mean Sum of square
et al.	:	and other workers
F_1	:	First filial generation
F_2	:	Second filial generation
Fe	:	Iron
g	:	Gram
GCV	:	Genotypic Coefficient of Variation
GA	:	Genetic Advance
GAM	:	Genetic Advance as a percentage of Mean
h	:	dominance
ha^{-1}	:	Per hectare
h^2_b or $h^2(b)$	:	Heritability in broad sense
h^2_n or $h^2(n)$	:	Heritability in narrow sense
i	:	Additive × additive
ICRISAT	:	International Crops Research Institute for Semi-Arid Tropics
j	:	Additive × dominance
K	:	Potassium
kg ha^{-1}	:	Kilograms per hectare
l	:	Dominance × dominance
LSD	:	Least Significant Difference
m	:	Mean
m ha	:	Million hectares

m t	:	Million tonnes
Mg	:	Magnesium
N	:	Nitrogen
No.	:	Number
P	:	Phosphorous
P_1	:	Parent 1
P_2	:	Parent 2
PCV	:	Phenotypic Coefficient of Variation
ppm	:	Parts per million
P_2O_5	:	Phosphorous pentoxide
r	:	Correlation coefficient
RBD	:	Randomized Block Design
S	:	Sulphur
S.Em	:	Standard error mean
viz.	:	Namely
WHO	:	World Health Organisation
ICP-OES	:	Inductively Coupled Plasma Optical Emission Spectrometry
Zn	:	Zinc

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ABSTRACT

The present investigation was carried out in sorghum to study the gene action involved in the inheritance of grain iron and zinc concentrations using generation mean analysis, to estimate the nature and magnitude of genetic effects and to understand the correlations of grain iron and zinc concentrations with grain yield and other agronomic traits. Two crosses (IS13205 × SPV1359 and IS13205 × IS23464) using diverse parents with varied levels of grain iron and zinc concentrations were made to generate six generations *i.e.*, P_1, P_2, F_1, F_2, B_1 and B_2. These six generations were evaluated in post-rainy season, 2012-13 at ICRISAT, Patancheru. Observations were recorded on important agronomic traits as per standard procedures, and grain iron and zinc concentrations were estimated using Inductively Coupled Plasma Optical Emission Spectometry (ICP-OES).

Analysis of variance showed significant differences among the generations of both the crosses for almost all the traits studied except days to 50% flowering, panicle width and 100-grain weight. Only one cross each showed significant difference between generations for days to 50% flowering (IS13205 × SPV1359) and 100-grain weight (IS13205 × IS23464). Panicle width exhibited non-significant difference between the generations of both the crosses. The phenotypic coefficient of variation was moderately higher than genotypic coefficient of variation for all the traits under study except days to 50% flowering, which suggested moderate influence of environment on these traits. Grain zinc concentration showed higher estimates of PCV % and GCV % than iron concentration in both the crosses and the gap between PCV % and GCV % was higher for grain zinc concentration than grain iron concentration which implies that the accumulation of this micronutrient suffered less from environment than that of grain zinc accumulation. High heritability coupled with high genetic advance was obtained for plant height, grain yield, grain iron and zinc concentrations indicating that these traits were governed by additive gene action and that selection will be effective. Average heterosis was negative for most of the traits

studied with significant negative heterosis over mid-parent obtained for panicle length and panicle width in the cross IS 13205 × IS 23464, significant average heterosis for grain yield which was negative in the cross IS 13205 × IS 23464 and positive in the cross IS13205 × SPV 1359 and was positive for iron and zinc concentrations in both the crosses. Significant residual heterosis over mid parent was obtained for plant height for both the crosses and for panicle width in the cross IS13205 × IS23464. Heterobeltiois was negative for almost all the traits studied. Significant negative heterobeltiosis obtained for grain yield for both the crosses, for days to 50 % flowering and plant height in the cross IS 13205 × IS 23464. Heterobeltiosis was non-significant for grain iron and zinc concentrations in both the crosses. Inbreeding depression was significant for plant height in the cross IS 13205 × IS 23464 and grain iron concentration in the cross IS13205 × SPV1359.

Correlation studies showed highly significant positive correlation between grain iron and zinc concentrations in both the crosses, indicating the possibility of simultaneous improvement of both the traits. Grain iron and zinc concentrations did not show any significant association with grain yield plant^{-1} and other agronomic traits except panicle length in the cross IS13205 × SPV1359.

Generation mean analysis showed that three scaling tests *viz.*, A, B and C were significant for all the traits studied, except for days to 50% flowering in the cross IS13205 × IS23464, with significance of atleast one scaling test (for grain yield and grain iron and zinc concentrations in both the crosses and for 100-grain weight in the cross IS13205 × SPV 1359) to three scaling tests (plant height and panicle length in the cross IS13205 × SPV 1359) which indicated the presence of digenic non-allelic interactions in the inheritance of these traits. For grain yield plant^{-1} both direct and epistatic effects were significant in the cross IS13205 × SPV 1359 whereas in the cross IS13205 × IS23464 additive and additive × additive were significant. For 100-grain weight, only additive × dominance interaction was significant which needs further investigation. For grain iron and zinc concentrations mostly digenic epistatic interactions were significant and direct effects were non-significant which indicated the predominance of epistatic interactions. In general, dominance component was higher in magnitude over additive component where as dominance × dominance component was of higher value than the remaining interactions, and also magnitudes of interactions were higher than their respective direct effects. Epistasis was duplicate type in all the cases except for plant height in the cross IS13205 × IS23464 where it was of complementary type. Recurrent selection, biparental mating and diallel selective mating system may be profitable to exploit both additive and non-additive components for bringing about improvement in grain yield and its attributes, grain iron and zinc concentrations.

Chapter I

INTRODUCTION

Chapter I

INTRODUCTION

Sorghum (*Sorghum bicolor* L. Moench) is a climate smart nutri-rich grain cereal and the fifth most important cereal crop in the world after wheat, maize, rice and barley. It is grown on 47 m. ha. in 104 countries, cultivated as staple cereal in Eritrea and Sudan, as a major food crop in much of South Asia and Africa, primarily to ease food security and in the USA, Australia and South America for animal feed. It is the fourth most important food crop in India after rice, wheat and pearl millet and is a staple food in central India. It is also the staple for large tribal populations across the country. India is one of the largest sorghum growers in the world with an area of 6.32 m.ha. and production of 6.03 m.t. with an average productivity of 954 kg ha^{-1}. In A.P., it covers an area of 0.25 m.ha. with a production of 0.44 m.t. with an average productivity of 1760 kg ha^{-1} (Indiastat, 2012).

Sorghum is the second cheapest source of energy (63.4 - 72.5% starch) and micronutrients such as iron (Fe) and zinc (Zn) after pearl millet. The poor and vulnerable groups in the society, particularly in India depend upon sorghum for their calorie and micronutrient requirement in the absence of access and affordability to nutrient-rich foods like vegetables, fruits and animal products. Earlier work at International Crops Research Institute for Semi Arid Tropics (ICRISAT) indicated that the contribution of sorghum in Fe intake was as high as 50% in the low income group population in India (Rao *et al*., 2006).

Producing nutritious and safe foods sufficiently and sustainably is the ultimate goal of modern agriculture. Past efforts were focussed on increasing crop yields, but enhancing the concentrations of mineral micronutrients has become an urgent task (Zhao and McGrath, 2009). Micronutrient malnutrition affects more than one-half of the world's population, especially women and preschool children (Nestel *et al.,* 2006). World Health Organization (WHO) of the United Nations estimated that under-nutrition is an underlying cause of 53% of all deaths in children under five years of age and recognized that the two micronutrients iron and zinc, and pro-vitamin A (β-carotene) are limiting (Ma *et al.,* 2008). It is estimated that, of the world's six billion people, 60-80% are iron deficient, and > 30% are zinc deficient (White and Broadley, 2006). An alternative solution is to increase mineral concentrations in edible crops.

This is termed 'Biofortification' (getting plants to do the work of fortificaion), which can be achieved by fertilization (Agronomic fortification), best traditional breeding practices and modern biotechnology (genetic biofortification). Biofortification is the method of enhancing grain micronutrient contents to combat the micronutrient (iron, zinc and vitamin A) malnutrition, also known as "hidden hunger", through genetic improvement.

To address this problem in 2004, Harvestplus, an international collaborative programme, was launched under the Consultative Group for International Agricultural Research (CGIAR) which is currently focussing on biofortificaiton for three micronutrients *viz*., kernel Fe, Zn and provitamin A in target crops (Pfeiffer and McClafferty, 2007). This programme was also launched at ICRISAT, Patancheru, to direct their research efforts towards improving the micronutrient contents of the grain in dryland cereals (sorghum and pearl millet) through biofortification. Biofortification, wherever possible, is a cost effective and sustainable solution for tackling the micronutrient deficiencies as the intake of micronutrients is on a continuing basis with no additional costs to the consumer in the developing countries (Ashok Kumar *et al*., 2011) which was ranked fifth important global issue to tackle within (as an alternative for the hidden hunger) by the Copenhagen Consensus Results (Meenakshi, 2007). Biofortification of sorghum by increasing mineral micronutrients, especially iron and zinc in the grains is of widespread interest (Pfeiffer and McClafferty, 2007 and Ashok Kumar *et al*., 2009).

The introduction of crop varieties selected and/or bred for increased Fe and Zn contents through plant breeding approach will complement the existing approaches (such as fortified foods and food supplementation while processing) to combat micronutrient deficiency. The plant breeding approach would avoid dependency on behavioural changes in farmers or consumers unlike other programs (Reddy *et al*., 2005). In order to realize the potential impact of the micronutrient-dense cultivars, the micronutrient-rich cultivars must be delivered in high-yielding backgrounds with farmer's preferred traits such as early maturity and large seed size (Ashok Kumar *et al*., 2010).

Preliminary studies at ICRISAT indicated limited variability for grain iron and zinc contents in hybrid parents, advanced breeding lines and released sorghum cultivars. The attempts to enhance the grain iron and zinc through exogenous application of micronutrients and macronutrients (ferti-fortification) did not increase the grain iron and zinc (Reddy *et al*., 2005). Hence, a genetic enhancement route was adopted to enhance the grain iron and zinc contents for which 29 accessions selected from core collection of the sorghum germplasm and breeding lines along with two check cultivars were evaluated. Results indicated a significant positive correlation between grain iron and zinc contents, and five accessions with high grain iron and zinc contents were identified (Ashok Kumar *et al*., 2009). Out of these four contrasting accessions (for iron and zinc contents) were crossed as parents in a diallele manner, which revealed that grain iron content was governed by additive gene action and grain zinc content was governed by additive gene action with little role of non-additive gene action (Gayathri, 2012). *There is no published report till date on detailed genetic dissection (through generation mean analysis) on kinds and magnitude of gene action controlling the grain iron and zinc concentrations in sorghum which show quantitative inheritance.* Hence, there is a need for further probe into the gene action to arrive at a sound conclusion and a breeding strategy.

The inheritance of quantitative traits is a moving target. The expression of these traits is affected not only by large number of genes governing them but also by environmental effects. Frequently, these genes interact with each other causing distortions in Mendelian ratios and leading to novel phenotypes (Phillips, 1998). The estimates of gene action in crop improvement programmes have direct bearing upon the choice of breeding procedures to be followed.

Phenotypic Coefficient of Variation and Genotypic Coefficient of Variation provides an idea about the range of variability present in the material used for the experiment. Heritability and genetic advance helps a breeder to know whether the selection will be effective or not in the improvement of a particular character. Estimates of heterosis are essential to know which of the selected parents will nick well to give rise to superior hybrids and the feasibility of release of hybrid as a commercial cultivar. Heterosis coupled with inbreeding depression reveals the gene action (additive or non-additive) involved in the inheritance of character. The correlation co-efficient provides a reliable measure of association among the characters and helps to differentiate vital associations useful in breeding from those of the non-vital ones (Falconer, 1981). Generation mean analysis (Hayman, 1958) gives a comprehensive picture of gene action controlling the trait based on first degree statistics.

Keeping this in view, the present investigation was undertaken with the following objectives:

- To study the gene action and heritability (narrow sense) for grain iron and zinc contents.
- To estimate correlation coefficients of yield and other characters with grain iron and zinc contents.

Chapter II

REVIEW OF LITERATURE

Chapter II

REVIEW OF LITERATURE

Sorghum is one of the important food and feed crops in the world which is cultivated in many parts of Asia and Africa, where its grains are used to make flat breads that form the staple food of many cultures. It is widely seen as poor man's crop because it is one of the main staple foods of the world's poorest and the most food insecure people. Presently it is cultivated in tropical, sub-tropical and temperate regions of the world extending in, as many as, six continents as 'great millet'. As it caters to the daily food intake needs of the poor, there is an absolute possibility to biofortify this crop to combat micro nutrient malnutrition, particularly iron and zinc. However, no much deep insight has been made into genetic architecture of grain iron and zinc concentrations in sorghum.

The choice of selection and breeding procedures for genetic improvement of any crop is largely dependent on the knowledge of type and relative amount of genetic components and the presence of non-allelic interactions for different traits in plant materials under investigation. Assessment of genetic effects involved in the expression of quantitative traits in sorghum can be accomplished by generation mean analysis. Generation mean analysis is a simple but useful technique for estimating genetic effects for a polygenic trait. Its greatest merit lies in its ability to estimate epistatic genetic effects such as additive × additive, dominance × dominance and additive × dominance. The information so obtained would have a direct bearing on the breeding programme for further tangible advancement of the crop. The correlation analysis identifies the traits which are positively or negatively associated with a particular trait. A brief review of literature available on above aspects in is presented in this section, under the following sub-headings:

- Variability, heritability and genetic advance
- Heterosis
- Inbreeding depression

- Correlation studies
- Generation mean analysis

2.1 Variability, heritability and genetic advance:

The information on the nature and magnitude of variability of different quantitative and qualitative traits in any crop species plays a vital role while formulating the efficient breeding programmes. Superior genotypes can be isolated by selection if considerable genetic variation exists within the population. Besides genetic variability, heritability and genetic advance also play a vital role in the improvement of any trait. The progress in breeding for the economic traits, which are mostly polygenically controlled and environmentally influenced, is determined by the nature and magnitude of their genotypic variability. Hence, it is essential to partition the overall variability into heritable and non-heritable components with the help of genetic parameters like genotypic and phenotypic coefficients of variation, heritability and genetic advance. The magnitude of heritability on one hand, and nature and extent of variability on the other hand gives an idea for effective genetic improvement through selection. The available literature on variability, heritability and genetic advance studies for all the characters studied in sorghum and for only grain iron and zinc concentrations in other cereal and millets, is furnished below:

Maloo *et al.* (1998) reported high potential genetic gain along with high estimates of GCA and broadsense heritability for quality traits like seed iron concentration in finger millet, indicating that this trait was largely governed by additive gene effects that in turn could be improved by selection.

Can and Yoshida (1999) obtained large proportion of phenotypic variance, which was attributable to the genotypic variance for plant height, 100-grain weight and grain yield in sorghum.

Lata and Sailesh (2001) reported that the estimates of GCV and PCV were higher for stover yield, biological yield and panicle weight among all the traits studied in 23 genotypes of sorghum in four environments.

Brkic *et al.* (2003) estimated the grain iron concentration in the range of 13.6-30.3 mg kg^{-1} and grain zinc concentration in the range of 16.0-23.6 mg kg^{-1} in maize and suggested the possibility to improve several minerals simultaneously due to significantly positive associations among minerals.

Oikeh *et al.* (2003) reported that kernel iron levels ranged from 16.8 to 24.4 mg kg^{-1}, while kernel zinc levels ranged from 16.5 to 24.6 mg kg^{-1} in maize. They further elaborated that genetic component accounted for 12% of the total variation in kernel iron and 29% for kernel zinc levels.

Kayode *et al.* (2006) determined that the iron concentration of the sorghum grains ranged from 30 to 113 mg kg^{-1} with an average of 58 mg kg^{-1} and the zinc concentration ranged from 11 to 44 mg kg^{-1} with an average of 25 mg kg^{-1}.

Oury *et al.* (2006) estimated iron and zinc concentrations in the range of 20 to 60 ppm and 15 to 35 ppm, respectively in bread wheat.

Reddy *et al.* (2006) reported considerable genetic variability coupled with high broad-sense heritability, suggesting good prospects of genetic enhancement for grain iron and zinc density through conventional breeding complemented by molecular breeding.

Bello *et al*. (2007) detected lower values of GCV compared to PCV for most of the traits studied with highest values recorded for yield $plot^{-1}$ and lowest values recorded for flag leaf width in sorghum.

Hemalatha *et al.* (2007) found that the variability in zinc concentration ranged from 1.08 mg 100 g^{-1} in rice to 2.24 mg 100 g^{-1} in sorghum and iron concentration ranged from 3.85 mg 100 g^{-1} in rice to 6.51 mg 100 g^{-1} in sorghum.

Warkad *et al*. (2008) observed highest PCV and GCV values for dry fodder yield $plant^{-1}$ followed by earhead breadth and length, grain yield $plant^{-1}$, stem girth and 1000 seed weight among the traits studied in sorghum.

Kusalkar *et al*. (2009) reported highest heritability in broad sense for growth traits, 1000-seed weight, grain yield, number of leaves per plant, leaf width, internode length, peduncle length and earhead length in sorghum. High heritability accompanied with high genetic advance was observed for growth traits *viz.*, grain yield, 1000-grain weight, number of leaves $plant^{-1}$, earhead length, leaf width, internode length, and peduncle length and plant height. Magnitude of PCV was found more than GCV for all traits.

Simic *et al*. (2009) found that there were significant genetic variations and workable heritabilities for Fe, Zn, P/Fe and P/Zn estimated in 294 F_4 lines of a maize population, but there were no associations among six traits according to both simple correlations and principal component analysis.

Becerra *et al*. (2010) reported that grain concentrations of iron and zinc varied from 27-86 and 39-115 mg kg^{-1}, respectively in *Triticum dicoccoides*. They further explained that accessions with higher nutrient concentrations had also greater grain weight, suggesting that higher grain iron and zinc concentrations were not necessarily related to small grain size or weight.

Chavan *et al*. (2010) recorded high PCV and GCV for harvest index (36.10) followed by grain yield $plant^{-1}$ (34.23), panicle width (27.67) in sorghum. The highest value of GCV

(25.49) and PCV (36.72) were observed for harvest index. The least values for PCV and GCV (27.10 and 5.97 respectively) were recorded for days to maturity.

Godharle *et al*. (2010) studied the genetic variability in 20 B lines and 16 R lines of *Kharif* sorghum and found that genotypic variance was lower than the phenotypic variance for all the traits studied in sorghum. High genotypic and phenotypic variance, heritability and genetic advance were observed for the traits panicle length, fodder yield, primary branches panicle^{-1}, grains primary branches^{-1}, harvest index, grain yield and plant height.

Ling and Kaun (2010) showed that the distribution of mineral element concentrations in the F_2 populations exceeded the performances of their parents, indicating the possibility for selecting the offspring with abundant mineral nutrients from the F_2 populations in rice.

Nguni *et al.* (2011) reported that farmer varieties of sorghum showed superiority for grain iron concentration ranging from 2.74 to 8.18 mg 100 g^{-1} and grain zinc concentration ranging from 2.03 to 5.53 mg 100 g^{-1}.

Samak *et al*. (2011) elaborated that PCV% and GCV% for grain iron and zinc were almost on par with each other in both F_4 and F_5 generations, derived from a cross in rice, indicating that the variation for these two traits was genetic in nature.

Govindaraj *et al*. (2011) recorded highest PCV% and GCV% for iron and moderate PCV% and GCV% for zinc among the grain micronutrients concentrations studied in pearlmillet. PCV% was greater than GCV% for almost all the traits studied which showed the influence of environmental factors on the traits studied.

Kamatar *et al*. (2011) observed high GCV, PCV and high heritability with high genetic advance for grain yield in both *kharif* × *rabi* and *rabi* × *rabi* progenies of sorghum.

Kumar *et al*. (2011) estimated genetic parameters for various quantitative traits of two interspecific crosses of sorghum and found that there was a close correspondence between PCV and GCV for all the traits studied except days to 50% flowering, panicle length and fodder yield of sorghum. High heritability coupled with high genetic advance was obtained for almost all traits except days to 50% flowering and fodder yield.

Langhi *et al*. (2011) detected higher phenotypic coefficient of variation than the genotypic coefficient of variation for all the traits studied. The highest genotypic coefficient of variation was observed for grain yield plant^{-1} and the lowest for days to maturity in sorghum.

Mahajan *et al*. (2011) obtained high values for PCV and GCV for harvest index followed by grain yield panicle^{-1}, panicle width and number of grains panicle^{-1}. High heritability coupled with high genetic advance was observed for number of grains panicle^{-1}, plant height and grain yield panicle^{-1} in sorghum.

Mohanraj *et al*. (2011) obtained higher values of PCV than GCV for all the traits studied. For grain yield plant^{-1} only one cross IS6354 × TNS 52 had high PCV and moderate GCV values coupled with high heritability and high GA as percent of mean in sorghum.

Shinde *et al*. (2011) observed that at both locations, derivatives of B × B and B × R exhibited higher PCV and GCV compared to R × R group for number of grains panicle^{-1} and grain yield plant^{-1} while R × R derivatives showed high PCV and GCV for these traits; number of primaries plant^{-1}, number of grains panicle^{-1} and grain yield plant^{-1} in sorghum.

Ali *et al*. (2012) obtained similar values of PCV and GCV for the traits studied with highest PCV and GCV recorded for panicle width in sorghum. Heritability in broad sense was high for all the traits studied with highest value obtained for grain yield plant $^{-1}$.

Chaudhary *et al*. (2012) reported that grain iron and zinc concentrations, plant height and grain yield per plant had high magnitude of phenotypic range, genotypic co-efficient of variation, phenotypic co-efficient of variation, heritability and genetic advance as per cent of mean, suggesting the importance of additive gene action in pearl millet.

Babu *et al*. (2012) found that the grain iron concentration showed large gap between PCV% (41.10) and GCV% (29.49) while for grain zinc concentration PCV% (11.74) and GCV% (10.88) correspond well with each other indicating greater and lesser influences of environment on these traits respectively in rice.

Seetharam and Ganeshmurthy (2013) reported similar values of PCV and GCV for all the agronomic traits studied with highest values obtained for panicle weight and single plant yield in sorghum. Number of leaves plant^{-1}, plant height and leaf width recorded high heritability coupled with high genetic advance.

2.2 Heterosis:

The superiority of F_1 hybrid over the parents is termed as heterosis. Heterosis works as a basic tool for improved production of crops in the form of F_1 hybrids. The phenomenon of heterosis of F_1 hybrids can reflect their own specific combining ability (SCA) and the general combining ability (GCA) of parental lines. Hence, heterotic studies can provide the basis for exploitation of valuable hybrid combinations and their commercial utilization in future breeding. Heterosis in sorghum had been widely exploited in the form of CSH series hybrids by various workers till today. A brief review of heterosis studies in sorghum is presented below:

Kirby and Atkins (1968) found that significant average heterosis was expressed by the sorghum hybrids for grain yield and plant height and also observed greatest heterotic response for grain yield, where hybrids averaged 122 % of the mid-parent values, with a range of 106 to 147 % among individual hybrids.

Rao and Goud (1977) concluded that the overdominance conditioned the grain yield in sorghum. They further observed maximum heterosis for grain yield in IS 2226 (dwarf introduction) × Karad Local (tall), resulting from additive × additive gene interaction.

Giriraj and Goud (1983) reported that heterosis for grain yield was mainly due to the heterotic effect expressed for 100-grain weight in sorghum which was not related to genetic divergence.

Nandanwankar (1990) concluded that sorghum hybrids showing significant heterobeltiosis for grain yield were also significantly superior for atleast 3 to 4 ear and grain traits and they further reported that heterosis for grain yield was due to increased number of seeds ear head^{-1}, secondaries and ear weight.

Badhe and Patil (1997) found that most of the heterotic combinations in sorghum were between geographically diverse parents and these were heterotic for all the panicle traits and plant height.

Rajguru *et al*. (2005a) identified the sorghum cross combinations 116A × RS-29, 3642A × SPV-489, 53A × SPV-1277 and 36642A × RSE-5 with the highest magnitude of heterosis for grain yield and its components.

Rajguru *et al*. (2005b) concluded that the *per se* performance of cytoplasmic genetic male sterile lines, restorer lines and their hybrids was related to heterotic effects in *rabi* sorghum.

Chaudhary *et al*. (2006) evaluated seven sorghum hybrids for their performance in *rabi* season and showed significant useful heterosis for grain yield and other yield contributing traits.

Desai *et al*. (2006) showed significant heterosis for all the yield components except panicle length in sorghum and further concluded that the crosses that exhibited heterobeltiosis showed higher grain size.

Iyanar and Gopalan (2006) found close association between *per se* performance of hybrids and heterosis for all the agronomic traits except for leaf number and seed yield, suggesting that the selection of the crosses based on *per se* performance could be more realistic in sorghum.

Jayalakshmi *et al*. (2006) identified three promising hybrids, i.e. NTJ 3 × CSV

13, NTJ 4 × NJ 2575 and NJT 4 × SPV 1532, with desirable heterobeltiosis for grain yield and panicle weight, which could be used for further exploitation in sorghum breeding programs.

Chen *et al*. (2007) showed that heterosis of iron concentration was very low, indicating that traditional hybrid breeding may not be efficient for improving this trait by analysing nine maize inbred lines.

Ghorade and Ghive (2007) reported highest heterobeltiosis (38.43%) in respect of grain yield, plant height, days to 50% flowering and all panicle traits in the sorghum cross MS-104A × AKR-354.

Khapre *et al*. (2007) concluded that the crosses SPV 1422 × SPV 1359, SPV 1500 × PVR 524, SPV 1502 × PVR 524, SPV 1422 × SPV 1413 and SPV 1500 × RR 9808 exhibited significantly high heterobeltiosis and standard heterosis over M 35¬1, CSH 15 R and SPV 655 in sorghum.

Kulkarni *et al*. (2007) reported pronounced hybrid vigour for grain yield, panicle length, panicle breadth, 1000-seed weight and fodder yield, while it was low for earliness, i.e. days to 50 % flowering in sorghum.

Vragolovic *et al*. (2007) noticed that heterosis was not significant for leaf and grain iron and zinc concentrations in maize. They further found that the iron and zinc concentrations varied from 15.9 to 24.8 mg kg^{-1} and 19.2 to 24.1 mg kg^{-1}, respectively.

Wadikar *et al*. (2007) identified the cross PMS 37 A × RS 29 with greatest significant heterotic effects for grain yield $plant^{-1}$ at both physiological and normal maturity in sorghum.

Rani and Rao (2009) found that there was no perfect correspondence between level of heterosis expressed in a cross and genetic divergence between parents in grain sorghum.

Makanda *et al*. (2010) found that the overall hybrid mean yield was significantly higher than that of parents and standard check varieties, which was attributed to high levels of average heterosis and standard heterosis, respectively in sorghum.

Showemimo *et al*. (2010) observed high heterotic value of 692.03% for grain yield in sorghum and concluded that the hybrids that exhibited high heterotic value for grain yield also had high heterotic value for yield components *viz*., 1000 seed weight and number of seed $panicle^{-1}$.

Sajjanar *et al*. (2011) reported that the cross, Atharga Kempu Jola × M 35-1 showed high level of significant positive heterosis for panicle weight (79.8 and 73.2%

over midparent and better parent/control, respectively) and grain yield (109.7 and 99.5% over midparent and better parent/control, respectively).

Velu *et al*. (2011) found that hybrids did not outperform the parents having high iron and zinc levels, which showed that there would be little opportunity, if any, to exploit heterosis for these mineral micronutrients in pearl millet.

Gayathri (2012) reported non-significant heterosis over better parent, mid parent and standard checks for both grain iron and zinc concentrations in sorghum which revealed the predominance of additive gene action.

Govindraj *et al*. (2013) observed not a single hybrid which could transgress better parent which indicated the lack of better parent heterosis for both iron and zinc concentrations in pearl millet. Midparent heterosis was significant for many hybrids mostly in negative direction for these traits.

Ashok Kumar *et al*. (2013) obtained significant positive heterosis over mid parent, better parent and standard check for iron concentration in many of the crosses studied compared to zinc concentration in sorghum. They further obtained highly significant positive correlation between mid parent value and hybrid performance *per se* for both iron and zinc.

In addition to the above studies, a brief review on heterosis for various traits in sorghum reported by several workers is presented hereunder:

Table 2.1. Heterosis for various traits in sorghum

Trait	Heterosis	References
Plant height	Positive heterosis over mid parent	Shivanna and Patil (1988) Belavatagi (1997)
	Significant heterosis over mid parent	Kanaka (1979)
	Non-significant heterosis over mid parental values.	Giriraj and Goud (1981)
	Significant negative heterosis over mid parent	Patil and Biaradar (2005)
	Positive heterosis over better parent	Senthil and Palaniswamy (1993) Ganesh *et al.* (1996) Lokapur (1997) Pawar (2000)
	Negative heterosis over	Naik *et al.* (1994)

	better Parent	Madhusudana and Patil (1996) Lokapur (1997) Pawar (2000)
	Heterosis over standard check	Shivanna and Patil (1988)
	Positive as well as negative Heterosis	Shivanna (1989)
Days to 50 % flowering	Heterosis over mid parent	Shivanna and Patil (1988) Belavatagi (1997)
	Positive heterosis over better parent	Senthil and Palaniswamy (1993) Pandit (1989)
	Negative heterosis	Naik *et al.* (1994) Lokapur (1997) Pawar (2000)

Table 2.1. (contd.)

Trait	Heterosis	references
100 grain weight	Significant positive heterosis over mid parent	Dinakar (1985) Sahib and Reddy (1986) Chinna and Phul (1988) Geeta and Rana (1988) Nandanwankar (1990) Patel *et al*. (1990) Senthil and Palaniswamy (1993) Sankarapandian *et al.* (1994) Lokapur (1997) Pawar (2000) Nimbalkar *et al*. (1988) Birdar (1995)
	Highly significant heterosis over mid-parent and better parent	Geeta and Rana (1988) Shivanna (1989) Senthil and Palaniswamy (1993) Biradar (1995) Chen (1994)

	Negative heterosis over mid-parent	Pandit (1989)
	Negative heterosis over better parent	Desai *et al.* (1985)
	Significant heterosis over mid parent, better parent and check.	Ganesh *et al.* (1996)

Table 2.1. (contd.)

Trait	Heterosis	References
Grain yield	Positive heterosis over mid parent	Nimbalkar *et al.* (1988) Wenzel (1988) Berenji (1988) Kasenko (1988) Chinna and Phul (1988)
	Positive heterosis over mid parent	Jeebaraj *et al.* (1988) Shivanna and Patil (1988) Nandanwankar (1990) Choudhari (1992) Rao *et al.* (1993) Sankarapandian *et al.* (1994) Ganesh *et al.* (1996)

A brief review of heterosis for grain iron and zinc reported by several workers in different crops is presented hereunder:

Table 2.2. Heterosis for grain iron and zinc concentrations in various crops

Trait	Crop	Heterosis	References
Grain iron	Pearl millet	Significant positive heterosis over mid-parent	Aruselvi *et al.* (2006) Velu (2006) Velu *et al.* (2011)
Grain zinc	Pearl millet	Significant positive heterosis over mid-parent	Aruselvi *et al.* (2006) Velu (2006) Velu *et al.* (2011)

2.3 Inbreeding depression:

The reduction in the vigour or size of the F_1 in succeeding segregating generations due to selfing or inbreeding is termed as inbreeding depression. Crosses between inbred lines show vigour and productiveness in F_1 generation (Shull, 1908), but with increasing homozygosity due to selfing, vigour and productiveness is reduced by 50% in each generation because of inbreeding depression (Falconer, 1981). The extent of inbreeding depression gives an idea about the productivity of hybrids, and also magnitude of transgressive segregants, which may be stabilized as improved inbreds. A brief review of inbreeding depression studies made by earlier workers in sorghum is presented below:

Rubencio and Romulo (1981) observed a great depression of F_2 generation of sorghum hybrids in terms of number of days to flowering, plant height at maturity, number of days to maturity, length of panicle, grain yield of sorghum, and weight of the 250 seeds.

Patel *et al*. (1983) obtained highly significant inbreeding depression for grain yield plant^{-1} in all hybrids. They further elaborated that the hybrids which showed maximum heterosis in F_1 did show maximum inbreeding depression in F_2 also for most of the traits under study. However in some cases though there was little heterosis, a marked inbreeding depression was observed.

Reddy and Joshi (1993) found that 1000 grain weight showed significant inbreeding depression in both heterotic and non-heterotic crosses. Also panicle length showed significant inbreeding depression for heterotic crosses in F_2 and in the later generation.

Rani *et al*. (2008) reported that inbreeding depression was significant for almost all traits studied in the four crosses except for leaf area index, relative water concentration and panicle length in two crosses, number of leaves plant^{-1} in three crosses and percent green leaves at maturity and panicle width in one cross.

2.4 Correlation studies:

The degree of association between two variables is measured by correlation coefficient which indicates the relationship between these two variables. It is the regression coefficient which measures the change in one variable for a unit change in other variable. Clear insight on genetic associations of iron and zinc concentration with grain yield and its contributing traits can help breeders in devising a suitable breeding strategy for enhancement of micronutrient density in sorghum grains. A review on the earlier research findings in this aspect is briefed below:

Rao *et al*. (1979) found that yield was negatively correlated with ash, magnesium and zinc. There was a negative association between days to maturity and zinc. He further concluded that the sorghum grain had a better balance of mineral composition as was evidenced by higher values of calcium, magnesium, phosphorous and iron than rice and maize.

Srihari and Nagur (1980) reported that grain yield of sorghum was significantly associated with 1000-grain weight and plant height.

Chintu *et al*. (1994) observed positive correlations of 1000 grain weight with iron (r = 0.80; P < 0.01) and zinc (r = 0.85; P < 0.01) concentration grain^{-1} which indicated that breeding for higher levels of these micronutrients could be achieved without compromising the large grain size in pearl millet.

Graham and Welch (1996) reported significant and fairly higher positive correlation (r = 0.55) between grain iron and zinc concentrations suggesting the possibility of combining both the micronutrients in single agronomic background. They further noted that the seeds rich in iron and zinc concentrations showed several agronomic advantages such as higher seedling vigor, especially in low- fertile soils, higher levels of resistance to diseases and empowering plants with higher water use efficiency.

Oikeh *et al*. (2003) reported that kernel iron was positively correlated with kernel zinc (R^2 = 0.51, P < 0.0001) in maize.

Reddy *et al*. (2005) reported significant and positive correlation between grain iron and zinc concentrations indicating the possibility of simultaneous improvement of both the traits in sorghum. Moreover they found a significant (negative) rather weaker correlation of grain iron concentration with grain and stover yields and relatively strong correlation of grain zinc concentration with grain and stover yields. The poor correlation of agronomic traits such as days to 50% flowering and plant height with grain iron and zinc concentrations indicated the possibility of developing micronutrient-dense lines in desired maturity and height background, with little compromise in grain and/or stover yields.

Velu (2006) reported negative association of grain iron (r = -0.60 to -0.31) and grain zinc (r = -0.32 to -0.53) concentrations with days to 50% flowering in pearl millet.

Morgunov *et al*. (2007) found a strong positive correlation between grain iron and zinc concentrations and negative but non-significant association of grain iron concentration (r = 0.05) with days to 50% flowering and 1000-grain weight, while significant negative correlation with grain yield (r = -0.41) in wheat. Similar correlations were found for zinc with other traits but the negative correlation(r= 0.64) of zinc concentration with the yield was stronger than that observed for iron concentration.

Montezano *et al*. (2008) determined that the linear correlation coefficients between nutrient concentration and grain yield were significant and negative for copper, manganese and zinc in maize.

Velu *et al*. (2008) reported that the correlation co-efficient between grain iron and zinc concentrations was positive and highly significant (r=0.66 to 0.85; P<0.01), indicating the likely effectiveness of simultaneous improvement for both micronutrients in pearl millet. He further found a negative but non-significant association of days to 50% flowering with grain iron (r = -0.29) and grain zinc (r = -0.31) concentrations.

Govindraj *et al*. (2009) found that the number of productive tillers, panicle length, panicle girth, 100-grain weight and days to maturity were significant and positively associated with grain yield in pearl millet. They further identified positive correlation between grain iron and zinc concentrations, suggesting simultaneous selection of these micronutrients.

Gupta *et al*. (2009) found significant positive correlation between grain iron and zinc concentrations (r=0.81 to 0.82; P<0.01) in pearl millet suggesting the possibility of simultaneous effective genetic improvement of both micronutrients. They further reported non-significant correlation of grain iron and zinc concentrations with grain yield and 1000-grain weight indicating that there would be no penalty on grain yield and seed size while breeding for grains rich in these micronutrients.

Ashok Kumar *et al*. (2009) observed significant positive correlation between grain iron and zinc concentrations, but a significant negative correlation of grain iron (r = -0.36) and zinc concentrations (r = -0.46) with grain yield in sorghum which was numerically low indicating that genetic enhancement for grain iron and zinc concentrations did not have yield penalty.

Zhao *et al*. (2009) found that grain zinc, but not iron, concentration correlated negatively with grain yield, and a decreasing trend in grain zinc concentration with the date of variety release, suggesting that genetic improvement in yield had resulted in a dilution of zinc concentration in grain. They further reported a positive correlation between grain zinc and iron concentrations in wheat.

Bello (2010) observed strong positive correlation between yield components and nutritive traits (protein, carbohydrate, ash, fat and fiber) of sorghum.

Chatzav *et al*. (2010) reported positive correlations among the concentrations of grain zinc, iron and protein in wheat, suggesting that all the three nutrients could be improved concurrently with no yield penalty.

Ashok Kumar *et al*. (2010) observed highly significant positive correlation between grain iron and zinc concentration ($r = 0.853$; $P<0.01$). They also observed weak association of grain iron and zinc concentrations with days to 50% flowering, plant height, grain yield and grain size indicating that there would be no penalty for enhancing grain iron and zinc concentrations in sorghum along with other agronomic traits such as grain size and grain yield in varied maturity backgrounds.

Na *et al*. (2010) reported a negative genetic correlation between micronutrient concentration and 1000-grain weight and positive phenotypic correlation between micronutrient concentration and grain yield of wheat and further stated the possibility for simultaneously improving grain concentrations of micronutrients, protein and yield in wheat.

Yong *et al*. (2010) reported that grain iron concentration was highly significant and positively correlated with that of grain zinc in wheat, indicating high possibility to combine high iron and zinc traits in wheat breeding. They further found strong positive correlations among the concentrations of iron, zinc and protein concentration.

Xian *et al*. (2010) observed significant and positive correlation between zinc and iron concentrations in wheat grain.

Anandan *et al*. (2011) observed negative correlation between grain yield and mineral concentrations and positive correlation among the mineral elements iron, zinc, manganese and copper in rice. Mineral element concentrations and cooking quality traits *viz*., kernel length after cooking and kernel linear elongation ratio were positively correlated, indicating the role of micronutrients in cooking quality traits. They concluded that that breeding for high minerals would be probably realistic.

Feng *et al*. (2011) reported the positive correlations among grain zinc, Iron and protein concentrations in wheat, suggesting that all these three traits were combinable and could be simultaneously enhanced.

Velu *et al*. (2011) found highly significant and positive correlation between grain zinc and iron concentrations ($r=0.81$; $P<0.01$) of pearl millet, indicating that simultaneous improvement of both the micronutrients would be effective.

Chakraborthi *et al*. (2011) reported that kernel iron and zinc concentrations were positively correlated, while no significant associations were found between either of the micronutrients with grain yield, suggesting the possibility of simultaneous improvement of both the kernel micronutrients without negatively impacting grain yield in the QPM hybrids.

Wang *et al.* (2011) found that the seed size showed no correlation with grain iron and zinc concentrations in wheat, while grain iron and zinc concentrations had significant negative correlation with grain yield.

Gayathri *et al*. (2012) reported that plant height and 100-grain weight had positive correlation with grain zinc, while days to 50% flowering showed negative association with grain zinc in the post-rainy seasons, 2010 and 2011. Grain yield showed positive association in 2010, while negative correlation in 2011 with grain zinc, which can be attributed to the influence of environment on these traits.

Nagesh *et al*. (2012) observed that grain yield was positively correlated with number of productive tillers plant^{-1}, test weight and number of grains panicle^{-1} in rice. A positive correlation between grain iron and zinc concentrations was also observed while there was no correlation between grain iron and zinc concentrations with grain yield.

Govindaraj *et al*. (2013) found highly significant and positive correlations between the iron and zinc densities and concluded that simultaneous selection for both micronutrients was likely to be effective with respective to these parameters.

2.6 Generation mean analysis:

The estimation of epistasis assumes more significance in view of the fact that in its presence, variance component estimates are likely to be biased hence inferences drawn from such estimates are more likely to be misleading. The magnitude of the bias depends upon the relative magnitude of epistatic effects comparatively to the deviations of additive (*d*) and dominance (*h*), type of prevailing epistasis and direction of dominance. The existence of large array of interactions in a polygenic system causes over-estimation of heritability (narrow sense) thereby causing an additional bias in predicted gains. Generation mean analysis is a powerful statistical procedure for detection of epistasis using several basic generations from a cross between two inbred lines. The review below throws some light on gene action studies carried out on yield contributing traits in sorghum and kernel iron and zinc concentrations in maize.

Raju (1979) reported that plant height and days to flowering in sorghum were under additive gene action. Epistasis was absent for days to flowering and plant height. Remaining traits are under dominance and the three epistatic interactions.

Nayakar (1973) observed that non-additive gene effects had a major contribution in the inheritance of plant height, 1000-grain weight and days to 50% flowering in sorghum. Grain yield was more influenced by dominance gene effects than other traits. Among interactions, additive × additive and dominance × dominance effects contributed towards heterotic effects of all the traits studied.

Indi and Goud (1981) reported the predominance of additive gene action for days to 50 % flowering, plant height, ear length, ear breadth and predominance of dominance gene action for rest of vegetative traits. None of the crosses showed predominance of additive gene action for number of primaries, ear weight and grain yield. They also highlighted the importance of epistatic interactions for many of the traits in sorghum.

Birdar and Borikar (1985) found that additive gene action and among interactions additive × additive and dominance × dominance were important in the inheritance of ear length in sorghum. For grain weight, both additive and dominance and all the three interactions were important.

Nimbalkar *et al*. (1986) through generation mean analysis found that panicle length and grain yield were under both additive and non-additive gene actions including epistatic interactions in sorghum. For 1000-grain weigh both additive and dominance were found significant, among interactions additive × additive and additive × dominance were found significant.

Rao *et al*. (1994) carried out generation mean analysis in six generations of two crosses involving an exotic (E 36-1), a local (M 35-1), and an improved variety (SPV 86) of *rabi* sorghum in respect of grain yield and its components which revealed that most of the traits studied in both crosses are under the control of dominance gene effect, followed by either additive × additive or dominance × dominance interactions with small proportion of additive gene effect except for 1000 grain weight where dominance × dominance was more important in cross E 36-1 × M 35-1 and dominance in cross E 36-1 × SPV 86.

Desai and Shukla (1995) reported that additive and dominance gene action is important for days to 50 % flowering, panicle breadth, and grain weight in sorghum. Dominance is predominant for panicle length and grain yield per plant. All the three epistatic interactions were important for grain yield plant^{-1}.

Audilakshmi and Aruna (2005) elaborated that dominance genetic effects along with additive × dominance and negative dominance × dominance interactions with duplicate epistasis were predominant in cross 463B × IS17600, but in cross AKMS 14B × IS 17600 dominance and additive gene effects and additive × dominance gene effects were important for grain size in sorghum. They further opined that F_2 plants segregated as 3:1 (round: flat) for grain shape and for grain luster they got a ratio of 9:7 (non-lustrous: lustrous).

Kaul *et al*. (2005) reported that significant estimates of scaling test indicated the presence of epistatic interaction for grain yield and test weight in the crosses. Additive × additive gene interaction was significant in all the crosses for grain yield and four crosses for test weight in sorghum. The dominance × dominance gene interactions for grain yield and test weight was non-significant.

Narain *et al*. (2007) observed that simple additive-dominance model was inadequate to explain the inheritance of yield and related traits in sorghum, reflecting the presence of epistatic interaction. All the four traits in majority of crosses were under the influence of dominance gene effects. Higher magnitude of dominance and dominance × dominance gene interactions could not be exploited in the crosses with duplicate epistasis as it minimizes the expression of heterosis.

Rani *et al*. (2008) observed importance of dominance and various interaction effects particularly additive × dominance type of epistasis for yield and panicle component traits in sorghum.

Chakraborti *et al*. (2010) reported higher magnitude of additive gene action as compared to dominance for kernel iron concentration, while dominance was relatively higher in case of kernel zinc concentration in maize. Additive × dominance component was significant in both the crosses studied for kernel iron concentration, whereas additive × additive component was predominant for kernel zinc concentration. Different non-additive components acted in opposite directions in both the crosses for kernel iron concentration which could be a bottle neck to exploit heterosis.

Chapter III

MATERIAL AND METHODS

Chapter III

MATERIAL AND METHODS

The present investigation was carried out at ICRISAT, Patancheru, Hyderabad, located at an altitude of 545m above mean sea level and at 17.53° N latitude and 78.27° E longitude, with an objective of deciphering the gene action involved in the inheritance of grain iron and zinc concentrations in sorghum during post-rainy season, 2012-13. The technique adopted in carrying out the present investigation to generate data and the statistical procedures followed for analyzing the data are described in the following sub-heads:

1. Generation of breeding material
2. Evaluation of experimental material
3. Statistical analysis

3.1 GENERATION OF BREEDING MATERIAL

The material comprised of six basic generations *viz.*, P_1, P_2, F_1, F_2, B_1 and B_2 derived from two crosses (IS13205 × SPV1359 and IS13205 × IS23464) involving three genotypes of sorghum with varied levels of iron and zinc. The details of the parental lines used in this experiment are furnished in Table 3.1.

Table 3.1. Details of parents with diversified levels of grain iron and zinc concentrations

Parental line	Pedigree / description	Grain iron concentration	Grain zinc concentration

		(mg kg^{-1})	(mg kg^{-1})
SPV 1359	RSLG 112-1-6	34.95	26.57
IS 23464	Caudatum land race from India	30.42	27.78
IS 13205	Guinea Caudatum land race from Spain	45.02	36.16

3.2 EVALUATION OF EXPERIMENATAL MATERIAL:

3.2.1 Development of F_2 and back cross (B_1 and B_2) generations:

Three parental lines, *viz*., IS13205, SPV1359 and IS23464 and two resultant F_1 hybrids (IS13205 × SPV1359 and IS13205 × IS23464), which were already developed, were planted in the field during post-rainy season, 2011-12 to produce F_2, B_1 and B_2 generations. F_2 generation of each cross was produced by selfing the F_1 plants while B_1 and B_2 generations were developed by crossing back each F_1 hybrids with their male (IS13205) and female (SPV1359 and IS23464) parents.

3.2.2 Field evaluation:

The six basic generations parents, *viz*., (P_1 and P_2), first and second filial generations (F_1 and F_2), first and second backcrosses (B_1 and B_2) of each combination of crosses were evaluated in a randomized complete block (RCB) design with three replications at ICRISAT, Patancheru, Hyderabad during post-rainy season, 2012-13 for grain iron and zinc concentrations. Each block comprised of two rows each of P_1, P_2 and F_1, four rows each of B_1 and B_2 and eight rows of F_2. Each row is of 2 m length with a spacing of 75 cm between rows and 10 cm between the plants making the plot size of 1.5 m^2. Two seeds were planted/hill with a spacing of 10 cm between the hills and thinned later to a single seedling/hill to obtain a population stand of nearly 20 plants/ plot in P_1, P_2 and F_1, 80 plants/plot in F_2 and 40 plants/plot in B_1 and B_2 each. The genotypes were control pollinated by selfing (by bagging the panicles before they shed the pollen/stigmas come out). The crop was supplied with a fertilizer dose of 80 kg N and 40 kg P_2O_5 per hectare and nitrogen was applied in three split doses. Hand weeding and earthing-up operations were practiced for weed control. The crop plants were irrigated as and when required during the cropping season till one week before maturity. Recommended and usual cultural practices were adopted to raise a good crop.

3.2.3 Observations recorded:

Observations on the following quantitative traits were recorded on 5 randomly selected plants in parents and F_1s and 50 randomly selected plants in F_2s and 10 randomly selected plants in B_1s and B_2s each for one replication.

3.2.3.1 Plant height (cm): Plant height was measured in centimeters by using a scale from base of the plant to the tip of the flag leaf in each hill at the time of harvest.

3.2.3.2 Days to 50% flowering: The numbers of days taken from the day of sowing to first flowering in 50% of plants were counted and recorded as days to 50% per cent flowering in each plot and in each replication.

3.2.3.3 Panicle length (cm): Panicle length was measured in centimeters from the base of the panicle to its tip using a scale.

3.2.3.4 Panicle width (cm): Panicle width was measured as the spread of primaries from the main rachis of the inflorescence from one side of the panicle to the other.

3.2.3.5 Plant aspect score: The agronomic desirability score was recorded on a scale of 1 to 5, where 1 = most desirable and 5 = least desirable was recorded in each plot and in each replication.

3.2.3.6 100-grain weight (g): One hundred well filled grains were counted at random from each plant of each generation and weighed after thorough drying to 12% moisture concentration and recorded in grams, with the help of electronic top pan balance (with precision of 0.01 g).

3.2.3.7 Grain yield plant^{-1} (g): The weight of filled grains harvested from the each plant was recorded in grams after drying the grains to the required moisture concentration (12 %).

3.2.3.8 Grain iron (Fe) and zinc (Zn) concentration (mg kg^{-1}): The panicles were harvested at maturity and the grain was threshed carefully without any contact with metal containers to avoid contamination. The cleaned seeds were collected in cloth bags and the grain iron and zinc concentrations in them was measured with Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES), the details of which were briefed below:

Principle of ICP-OES:

ICP, abbreviation for Inductively Coupled Plasma, is one method of optical emission spectrometry. When plasma energy is given to an analysis sample from outside, the component elements (atoms) are excited. When the excited atoms return to low energy position, emission rays (spectrum rays) are released and the emission rays that correspond to the photon wavelength are measured. The element type is determined based on the position of the photon rays, and the concentration of each element is determined based on the rays' intensity. To generate plasma, first, argon gas is supplied to torch coil, and high frequency electric current is applied to the work coil at the tip of the torch tube. Using the electromagnetic field created in the torch tube by the high frequency current, argon gas is ionized and plasma is generated. This plasma has high electron density and temperature (10000K) and this energy is used in the excitation-emission of the sample. Solution samples are introduced into the plasma in an atomized state through the narrow tube in the center of the torch tube.

ICP-OES determination:

Determinations were carried out using A Prodigy High Dispersion Inductively Coupled Plasma (ICP) Spectrometer equipped with a dual view torch and 60 position auto sampler. The Prodigy is a compact bench-top simultaneous ICP-OES featuring an 800 mm focal length Echelle optical system coupled with a mega-pixel Large Format Programmable Array Detector (L-PAD). At 28 × 28 mm, the active area of the L-PAD is significantly larger than any other solid-state detector currently used for ICP-OES. This combination allows Prodigy to achieve significantly higher optical resolution than other solid-state detector based ICP systems. The detector also provides continuous wavelength coverage from 165 to 1100 nm permitting measurement over the entire ICP spectrum in a single reading without sacrificing wavelength range or resolution. This detector design is inherently anti-blooming and is capable of random access, non–destructive readout that results in a dynamic range of more than 6 orders of magnitude.

3.3 STATISTICAL ANALYSIS

The data recorded on different traits were subjected to the following statistical analysis.

1. Analysis of variance

2. Estimation of genetic parameters like genotypic and phenotypic coefficients of variation, heterosis, residual heterosis, inbreeding depression, degree of dominance, heritability in narrow sense and broad sense and genetic advance under selection and genetic advance as percent of mean

3. Estimation of correlation coefficients

4. Estimation of gene effects by Generation Mean Analysis

   a. Scaling tests of Mather (1949)

   b. Six parameter model of Hayman (1958)

3.3.1 Analysis of Variance

Before proceeding to the biometrical genetic analysis of the data, the genotypic differences among the entries were tested. Further analysis was carried out only when the mean squares due to genotypes were significant. Hence the data for different traits were statistically analyzed for significance by using analysis of variance (ANOVA) as described by Panse and Sukhatme (1985). The adopted design was Randomized Block Design (RBD) replicated thrice.

The linear mathematical model:

$$Y_{ij} = m + g_i + r_j + e_{ij}$$

Where,

Y_{ij} = Phenotypic observation of the i^{th} genotype in the j^{th} replication

m = General mean

g_i = Effect of i^{th} genotype

r_j = Effect of j^{th} replication

e_{ij} = Random error associated with i^{th} genotype in the j^{th} replication

The analysis of variance (ANOVA) was carried out for each trait as indicated below

ANOVA			
Source	**d.f.**	**MS**	**F calculated**

Replications (r)	(r-l)	Mr	Mr / Me
Treatments (t)	(t-l)	Mt	Mt / Me
Error (e)	(r-l) (t-l)	Me	
Total	(rt-l)		

Where,

r = number of replications

t = number of treatments (genotypes)

d.f. = degrees of freedom

MS = mean sum of squares

Mr = mean sum of squares due to replications

Mt = mean sum of squares due to treatments

Me = mean sum of squares due to error

The significance of mean sum of squares for each trait was tested against the corresponding error degrees of freedom using 'F' test (Fisher and Yates, 1967).

Estimation of mean values, standard error, critical difference and coefficient of variation (analysis of means)

Mean values were calculated as averages over replications for each respective treatments. The comparison between any two treatment means was performed by computing least significant difference (L.S.D.) or critical difference (C.D.) at 0.05 to 0.01 level of probability. The difference between any two treatment means was recorded as significant if they were greater than C.D.

$$\text{C.D.} = \text{S.E.} \times t\ edf$$

$$= \frac{(\sqrt{2\ Me})}{r} \times t\ edf$$

Where,

S.E. = standard error of the difference of the treatment means to be compared

$$= \underline{(\sqrt{2\ Me})} \times t\ edf$$

r

t_{edf} = Student's tabulated value at 5% and 1% level of significance at error degrees of freedom for the error mean square

Coefficient of Variation (C. V.)

The coefficient of variation is a unit less measurement and is used for comparing the extent of variation between different traits with different scales. It is the ratio of the standard deviation to the grand mean or experimental mean of the trait.

$$C.V. = \frac{\sqrt{Me}}{\overline{X}}$$

where,

$\overline{X}$ = Grand mean

3.3.2 Estimation of components of variances and genetic parameters

3.3.2.1 Estimation of components of variances

From the analysis of variance table, the mean sum of squares between genotypes will consist of the variances (i) due to genotypic differences and (ii) due to the environmental variation among the individuals of each genotype. Therefore, the genotypic and phenotypic variances were calculated as per the formula given by Burton and Devane (1953).

Environmental variance, $\sigma^2_e = Me$

Genotypic variance, $\sigma^2_g = \frac{Mt - Me}{r}$

Phenotypic variance, $\sigma^2_p = \sigma^2_g + \sigma^2_e$

3.3.2.2 Estimation of genetic parameters

3.3.2.2.1 Phenotypic and genotypic co-efficients of variation

The components of variances were used to estimate genetic parameters like phenotypic and genotypic co-efficients of variation (PCV and GCV) as per the formulae given by Falconer (1981).

Phenotypic standard deviation, $(\sigma_p) = \sqrt{\sigma^2_p}$

$$= \sqrt{\sigma^2_g + \sigma^2_e}$$

$$\text{PCV (\%)} = \frac{\text{Phenotypic standard deviation } (\sigma_p) \times 100}{\text{Grand mean } (\overline{X})}$$

$$\text{Genotypic standard deviation, } (\sigma_g) = \sqrt{\sigma^2_g}$$

$$\text{GCV (\%)} = \frac{\text{Genotypic standard deviation } (\sigma_g) \times 100}{\text{Grand mean } (\overline{X})}$$

Categorization of the range of variation was followed as reported by Subramanian and Menon (1973).

Low	:	Less than 10%
Moderate	:	10 to 20%
High	:	More than 20%

3.3.2.2.2 Heterosis:

Heterosis was estimated for two hybrids for eight traits using the following formulae. Estimates of heterosis were calculated according to Fonseca and Patterson (1968).

3.3.2.2.2.1 Heterosis over mid-parent (Relative Heterosis)

Heterosis was expressed as per cent increase or decrease observed in the F_1 over the mid-parent as per the following formula.

$$\text{Relative heterosis (\%) } (H_1) = \frac{\overline{F_1} - \overline{MP}}{\overline{MP}} \times 100$$

Where,

$\overline{F_1}$ = Mean of F_1

$\overline{MP}$ = Mean of parents

3.3.2.2.2.2Residual heterosis over mid-parent

The heterosis over mid-parent or average/relative heterosis present in F_2 generation is calculated as residual heterosis over mid-parent. It is calculated as follows (Rao, 1980) :

Residual relative heterosis (%) (H_1) = $\frac{\overline{F}_2 - \overline{MP}}{\overline{MP}} \times 100$

3. 3.2.2.2.3 Heterosis over better parent (Heterobeltiosis)

Heterobeltiosis was expressed as per cent increase or decrease observed in F_1 over the better parent as per the formula of Liang *et al*. (1971).

Heterobeltiosis (%) (H_2) = $\frac{\overline{F}_1 - \overline{BP}}{\overline{BP}} \times 100$

Where,

$\overline{\text{BP}}$ = Mean of better parent

(For the traits like days to 50% flowering, earliness is desirable so the early parents are taken as better parents).

3.3.2.2.2.4 Residual heterosis over better parent

The heterosis over better parent or heterobeltiosis present in F_2 generation is calculated as residual heterosis over better parent. It is calculated as follows (Rao, 1980):

Residual heterosis over better parent (%) (H_2) = $\frac{\overline{F}_2 - \overline{BP}}{\overline{BP}} \times 100$

The significance of heterosis, was tested by using 't' test as suggested by Snedecor and Cochran (1989) and Paschal and Wilcox (1975).

Where,

$$\text{Heterosis } t = \frac{\overline{\text{F}}_1 - \overline{\text{MP}}}{\sqrt{1.5\text{EMS}/\text{r}}}$$

EMS = Error Mean sum of Square

$$\text{Heterosis } t = \frac{F_1 - BP}{\sqrt{2\ EMS/r}}$$

r = Number of replications

The calculated ‘t’ value was compared with table ‘t’ value at error degrees of freedom.

3.3.2.2.3 Inbreeding depression

The loss of fitness in the progenies or decline in trait expression with decreased heterozygosity arising from consanguineous mating is known as inbreeding depression or inbreeding decline. It can be calculated using the following formula given by Kempthrone (1957):

$$\text{Inbreeding depression} = \frac{\overline{F_1} - \overline{F_2}}{\overline{F_1}} \times 100$$

Where $\overline{F_2}$ = mean of F_2 population

$\overline{F_1}$ = mean of F_1

The significance of inbreeding depression was tested by using ‘t’ test as indicated in the heterosis.

3.3.2.2.4 Degree of dominance

The magnitude of variance due to dominance deviations, relative to that of additive genetic variance (Robinson *et al.* 1949) expressed as a square root of ratio of additive variance to dominance variance is known as degree of dominance. It is calculated as follows:

$$\text{Degree of dominance} = \sqrt{H/D}$$

3.3.2.2.5 Heritability in broad sense:

It is the ratio of genotypic variance to the phenotypic variance. It is the heritable portion of phenotypic variance which responds to selection. It can be calculated using the formula (Hanson *et al.*, 1956):

$$h^2\ (b) = \frac{\text{genotypic variance } (\sigma^2 g)}{\text{Phenotypic variance } (\sigma^2 p)} \times 100$$

3.3.2.2.6 Heritability in narrow sense [h^2 (n)]

Heritability in the narrow sense refers to the proportion of additive variance to the total observed variance in the total population. Heritability in narrow sense [h^2 (n)] was calculated according to the formula given by Allard (1960) and expressed in percentage.

$$h^2 (n) = \frac{\text{Additive variance } (\sigma^2 a)}{\text{Phenotypic variance } (\sigma^2 p)} \times 100$$

The range of heritability estimates were categorized as follows as suggested by Johnson *et al.* (1955):

Low	:	0-30%
Medium	:	> 30-60%
High	:	> 60%

3.3.2.2.7 Genetic advance (GA)

Genetic advance refers to the expected genetic gain or improvement in the next generation by selecting superior individuals under certain amount of selection pressure. From the heritability estimates, the genetic advance was estimated by the following formula given by Burton (1952):

$$GA = K.\ h^2 (b).\ \sigma_p$$

Where,

GA	=	Expected genetic advance
K	=	Selection differential, the value of which is 2.06 at 5% selection intensity
σ_p	=	Phenotypic standard deviation
h^2 (b)	=	Heritability in broad sense

3.3.2.2.8 Genetic advance as percent of mean (GAM)

In order to visualize the relative utility of genetic advance among the traits, genetic advance as percent for mean (GAM) was computed as described by Johnson *et al*. (1955)

$$GAM = \frac{GA \times 100}{\text{Grand mean, } (\bar{X})}$$

The range of genetic advance as percent of mean was classified as suggested by Johnson *et al*. (1955).

Low	:	< 10 %
Moderate	:	10-20 %
High	:	> 20 %

3.3.4 Estimation of Simple Correlation Coefficients

Trait association refers to the association of traits *i.e.*, a change in one trait is accompanied by a change in the other trait. The data of eight traits were utilized for the computation of correlation coefficients between grain iron and zinc concentrations with other traits for all the genotypes. The formulae suggested by Snedecor and Cochran (1967) were utilized for the computation of correlation coefficients.

$$r(xy) = \frac{\text{Cov}(xy)}{\sqrt{(\text{Var } x)\ (\text{Var } y)}}$$

$$Cov(xy) = \frac{1}{n}\left[\sum xy - \frac{(\sum x \sum y)}{n}\right]$$

$$Var(x) = \frac{1}{n}\left[\sum x^2 - \frac{(\sum x)^2}{n}\right]$$

Where,

r(xy)	=	Correlation between x and y
Cov(xy)	=	Covariance for traits x and y
Var(x)	=	Variance for x
Var(y)	=	Variance for y
r	=	Correlation coefficient
xy	=	Two independent variables

To test the significance of correlation coefficients, the estimated values were compared with the table values of correlation coefficients (Fisher and Yates, 1967) at

5% and 1% levels of significance with (n-2) degrees of freedom, where ‘n’ is the total number of observations used.

3.3.5 Generation Mean Analysis

The concept of Generation Mean Analysis (GMA) was developed by Hayman (1958) and Jinks and Jones (1958) for the estimation of genetic components of variation. This technique involves six different generations *viz.*, parents (P_1 and P_2), their F_1, F_2 and back crosses (B_1 and B_2). Accordingly, the means were computed for each generation of P_1, P_2, F_1, F_2, B_1 and B_2 for each cross. The variance and corresponding standard errors of the means were computed from the deviations of the individual values obtained from individual plants for each of the generation in each cross and were analyzed to estimate various genetic parameters and the type of gene action involved in the inheritance of various traits.

The biometrical analysis consists of two main steps, *viz.*, (i) testing for epistasis and (ii) estimation of gene effects and variances.

3.3.5.1 Scaling test

The test which provides information regarding presence / absence of gene interaction is termed as scaling test. The test of adequacy of scales is important because in most of the cases the estimation of additive and dominance components of variances were made assuming the absence of gene interactions. Mather (1949) and Hayman and Mather (1955) gave four scaling tests to test the adequacy of additive-dominance model. The different scales, variances and standard errors are computed by using the following formulae:

$$\text{Scale A} = 2\overline{B_1} - \overline{P_1} - \overline{F_1} = 0$$

$$\text{Scale B} = 2\overline{B_2} - \overline{P_2} - \overline{F_1} = 0$$

$$\text{Scale C} = 4\overline{F_2} - 2\overline{F_1} - \overline{P_1} - \overline{P_2} = 0$$

Where,

P_1, P_2, F_1, F_2, B_1 and B_2 are means of different generations over the replications.

The variances of the quantities A, B, C and D were calculated from respective variances of different generations as follows

$$V_A = 4V(\overline{B_1}) + V(\overline{P_1}) + V(\overline{F_1}) = 0$$

$$V_B = 4V(\overline{B_2}) + V(\overline{P_2}) + V(\overline{F_1}) = 0$$

$$V_C = 16V(\overline{F_2}) + 4V(\overline{F_1}) + V(\overline{P_1}) + V(\overline{P_2}) = 0$$

$$V_D = 16\,V\overline{F_3} + 4V\overline{F_2} + V\overline{P_1} + V\overline{P_2} = 0$$

Where, V_A, V_B, V_C and V_D are the variances of the scale A, B, C and D; $V\overline{P_1}$, $V\overline{P_2}$, $V\overline{F_1}$, $V\overline{F_2}$, $V\overline{B_1}$ and $V\overline{B_2}$ are the variances of means of P_1, P_2, F_1, F_2, B_1 and B_2 generations, respectively. The variance of mean for each generation was calculated by dividing variance obtained from ANOVA table by the number of observations taken. The standard error of the scale A, B, C and D were worked out by taking the square root of respective variances.

$$\text{S.E. A} = \sqrt{V_A}$$

$$\text{S.E. B} = \sqrt{V_B}$$

$$\text{S.E. C} = \sqrt{V_C}$$

$$\text{S.E. D} = \sqrt{V_D}$$

The ‘t’ values were calculated by dividing the scale effects of A, B, C and D by their respective standard error.

t cal for A-test = Scale A / S.E. A

t cal for B-test = Scale B / S.E. B

t cal for C-test = Scale C / S.E. C

t cal for D-test = Scale D/ S.E. D

The calculated values of ‘t’ were compared with ‘t’ table values at 5% and 1% level of significance at their respective degrees of freedom. In each test, the degrees of freedom was taken as the sum of the degrees of freedom of various generations involved in that scaling test and the degrees of freedom for any generation was calculated as total number of observations minus number of replications. However, in case of unreplicated data the degrees of freedom will be number of observations per generation minus one.

If the calculated value of these scales is higher than ‘t’ table values (when *d.f.* = > 30, then ‘t’ table values are 1.96 and 2.58 at 0.05 and 0.01 level of probability), it is considered significant and vice versa. The significance of any one of these scaling tests indicates the presence of epistasis. It is to be noted that,

(a) D provides a test largely of ‘*i*’ type of interaction (additive × additive).

(b) C indicates ‘*l*’ (dominance × dominance) type of gene interaction.

(c) Significance of C + D relates to ‘*i*’ (additive × additive) and ‘*l*’ (dominance × dominance) type of interaction.

(d) ‘*j*’ (additive × dominance) type of interaction has no effect on C and D but it affects A and B. A and B tests provide evidence on *i*, *j* and *l* type of gene interactions (Singh and Chaudhary, 1977).

3.3.5.2 Components of Generation Means

When the scales were significant, the mean values over replication were used for the estimation of the gene effects. Owing to the presence of six generations (P_1, P_2, F_1, F_2, B_1 and B_2) in each cross, six parameter model proposed by Hayman (1958) and Jinks and Jones (1958) was followed to estimate the genetic parameters *viz.*, mean (*m*), additive gene effects (*d*), dominance gene effects (*h*) and three types of non-allelic gene interactions *viz.*, additive × additive (*i*), additive × dominance (*j*) and dominance × dominance (*l*).

Estimation of genetic parameters by six parameter model (Hayman, 1958)

m = Mean = $\overline{F}_2$

d = Additive effect = $\overline{B}_1 - \overline{B}_2$

h = Dominance effect = $\overline{F}_1 - 4\overline{F}_2 - (1/2)\overline{P}_1 - (1/2)\overline{P}_2 + 2\overline{B}_1 + 2\overline{B}_2$

i = Additive × Additive effect = $2\overline{B}_1 + 2\overline{B}_2 - 4\overline{F}_2$

j = Additive × Dominance effect = $\overline{B}_1 - (1/2)\overline{P}_1 - \overline{B}_2 + (1/2)\overline{P}_2$

l = Dominance × Dominance effect = $\overline{P}_1 + \overline{P}_2 + 2\overline{F}_1 + 4\overline{F}_2 - 4\overline{B}_1 - 4\overline{B}_2$

Where,

$\overline{P}_1$, $\overline{P}_2$, $\overline{F}_1$, $\overline{F}_2$, $\overline{B}_1$ and $\overline{B}_2$ are the mean values of P_1, P_2, F_1, F_2, B_1 and B_2 generations, respectively.

3.3.5.3 Test of significance of various gene effects

The test of significance of the gene effects was done by 't' test for which variance and standard error of each estimates were calculated using following equations.

Calculation of variances:

$V_m = V(\overline{F}_2)$

$V_d = V(\overline{B}_1) + V(\overline{B}_2)$

$V_h = V(\overline{F}_1) + 16V(\overline{F}_2) + (1/4)V(\overline{P}_1) + (1/4)V(\overline{P}_2) + 4V(\overline{B}_1) + 4V(\overline{B}_2)$

$V_i = 4V(\overline{B}_1) + 4V(\overline{B}_2) + 16V(\overline{F}_2)$

$V_j = V(\overline{B}_1) + 1/4V(\overline{P}_1) + V(\overline{B}_2) + 1/4\ V(\overline{P}_2)$

$V_l = V(\overline{P}_1) + V(\overline{P}_2) + 4V(\overline{F}_1) + 16V(\overline{F}_2) + 16V(\overline{B}_1) + 16V(\overline{B}_2)$

$V(\overline{P}_1)$, $V(\overline{P}_2)$, $V(\overline{F}_1)$, $V(\overline{F}_2)$, $V(\overline{B}_1)$ and $V(\overline{B}_2)$ were the variances of P_1, P_2, F_1, F_2, B_1 and B_2 generations, respectively.

Calculation of standard error:

The standard error of each of the gene effects was estimated as follows

S.E. (m) = $\sqrt{V_m}$

S.E. (d) = $\sqrt{V_d}$

S.E. (h) = $\sqrt{V_h}$

S.E. (i) = $\sqrt{V_i}$

S.E. (j) = $\sqrt{V_j}$

S.E. (l) = $\sqrt{V_l}$

The 't' values were worked out using following formulae

t (m) = m / S.E. (m)

t (d) = d / S.E. (d)

$$t(h) = h / S.E.(h)$$

$$t(i) = i / S.E.(i)$$

$$t(j) = j / S.E.(j)$$

$$t(l) = l / S.E.(l)$$

The significance for above genetic parameters were tested with the help of ‘t’ test. First standard error (S.E.) is worked out for each component separately by taking the square root of the variance of the respective component. Significance of the genetic effects is tested using ‘t’ test in a similar manner as in the case of scaling test. If the calculated value is greater than ‘t’ table value, it is considered significant and vice versa (Singh and Chaudhary, 2001).

Chapter IV

RESULTS AND DISCUSSION

Chapter IV

RESULTS AND DISCUSSION

The present investigation was carried out in sorghum on generation mean analysis using six generations of two crosses developed by involving the parents with contrasting grain iron and zinc concentrations for each cross. The results obtained are detailed here under the following headings:

4.1 Analysis of variance

4.2 Mean performance

4.3 Various genetic parameters

4.4 Heterosis and inbreeding depression

4.5 Correlation studies

4.6 Gene effects

4.1 ANALYSIS OF VARIANCE

The analysis of variance of two crosses for various agronomic traits and grain iron and zinc concentrations are presented in Table 4.1. The mean sum of squares due to generations was significant for both the crosses for the traits plant height, panicle length, grain yield plant^{-1}, plant aspect score and grain iron and zinc concentrations which revealed the existence of genetic diversity in the material under study for these attributes. For days to 50% flowering and 100-grain weight, mean sum of squares due to generations were significant only in one cross each *i.e.*, IS13205 × IS23464 and IS13205 × SPV1359, respectively. The mean sum of squares due to generations was non-significant for panicle width in both the crosses. The data pertaining to traits that exhibited non-significant deviation were not subjected to generation mean analysis.

4.2 MEAN PERFORMANCE

The *per se* performance of six generations (P_1, P_2, F_1, F_2, B_1 and B_2) of two crosses for various agronomic traits and for grain iron and zinc concentrations are furnished in Table 4.2. The female and male parents are indicated as P_1 and P_2. The results obtained are discussed trait-wise for each cross below:

Table 4.1. Analysis of variance (mean squares) of generations for various agronomic traits and grain iron and zinc concentrations in two crosses of sorghum during post-rainy season, 2012-13

Trait/cross	**Mean sum of squares**		
	Replications	**Generations**	**Error**
Degrees of freedom	2	5	10
IS13205 x SPV1359			
Days to 50% flowering	20.667	69.033	61.000
Plant height (cm)	82.698	3551.062**	192.139
Panicle length (cm)	2.700	32.515**	1.407
Panicle width (cm)	0.912	1.321	1.021
Grain yield plant^{-1} (g)	31.771	1664.975**	166.408
Plant aspect score	0.055	0.889**	0.255
100-grain weight (g)	0.487	0.684**	0.089
Grain iron concentration (mg kg^{-1})	0.382	139.271**	7.146
Grain zinc concentration (mg kg^{-1})	24.549	87.681**	13.170
IS13205 x IS23464			
Days to 50% flowering	45.389	134.056**	17.856
Plant height (cm)	80.733	2636.847**	148.484
Panicle length (cm)	9.847	17.016**	2.510
Panicle width (cm)	0.786	3.536	1.340
Grain yield plant^{-1} (g)	171.779	1036.557*	218.913
Plant aspect score	0.167	0.633**	0.100
100-grain weight (g)	0.174	0.300	0.128
Grain iron concentration (mg kg^{-1})	7.105	96.504**	11.010
Grain zinc concentration (mg kg^{-1})	14.727	111.366**	12.169

***** – Significant at 5% level of probability

** – Significant at 1% level of probability

4.2.1 Days to 50% flowering

The analysis of variance (Table 4.1) revealed non-significant difference between the generations for mean days to 50% flowering in the cross IS13205 × SPV1359, since both the parents had same duration of days to 50% flowering (70.33). Hence, the data were not subjected to generation mean analysis. The low variability observed for this trait among the six generations indicated negligible contribution of this character to total variability.

In the cross IS13205 × IS23464, there was a significant difference between the parents for mean days to 50% flowering and the female parent P_1 (63.33) flowered earlier compared to male parent P_2 (79.67). The mean of F_1 (66.33) and F_2 (65.67) were on a par with the early parent P_1, but were significantly lower than the late parent P_2. In case of back crosses, B_1 (62.00) flowered earlier than the early parent P_1 and B_2 flowered earlier compared to late parent P_2. B_1 flowered earlier compared to B_2 (Fig. 4.1).

4.2.2 Plant height (cm)

Both the parents differed significantly from each other (Table 4.2) and the parent P_2 (248.75) recorded higher plant height than P_1 (173.05) in the cross IS13205 × SPV1359. The hybrid F_1 (230.83) expressed greater average plant height in comparison to mid-parent, but was lower than the better parent. Further, the mean performance of F_2 (234.74) was close to F_1 and was also lower than the better parent like the F_1. Among the back crosses, B_2 (179.61) recorded more plant height than B_1 (176.41) and was close to parent P_1.

In the cross IS13205 × IS23464, the plant height was significantly different in both the parents (Table 4.2) and the parent P_2 (249.18) recorded higher plant height than P_1 (181.10). The hybrid F_1 (216.88) recorded slightly higher plant height than mid parent value but it remained shorter than better parent. The F_2 (180.47) generation showed plant height similar to that of shorter parent P_1. Among the back crosses, significant differences were observed between B_1 (195.78) and B_2 (169.28) and B_1 stood higher in plant height than B_2 which recorded lesser height compared to, even the shorter parent P_1 (Fig. 4.1).

4.2.3 Panicle length (cm)

The data from Table 4.2 revealed that in the cross IS13205 × SPV1359 parent P_1 (22.33) recorded slightly higher panicle length than the parent P_2 (21.08). The mean performance of F_1 (22.11) was almost on a par with the parent P_1. F_2 (22.95) generation

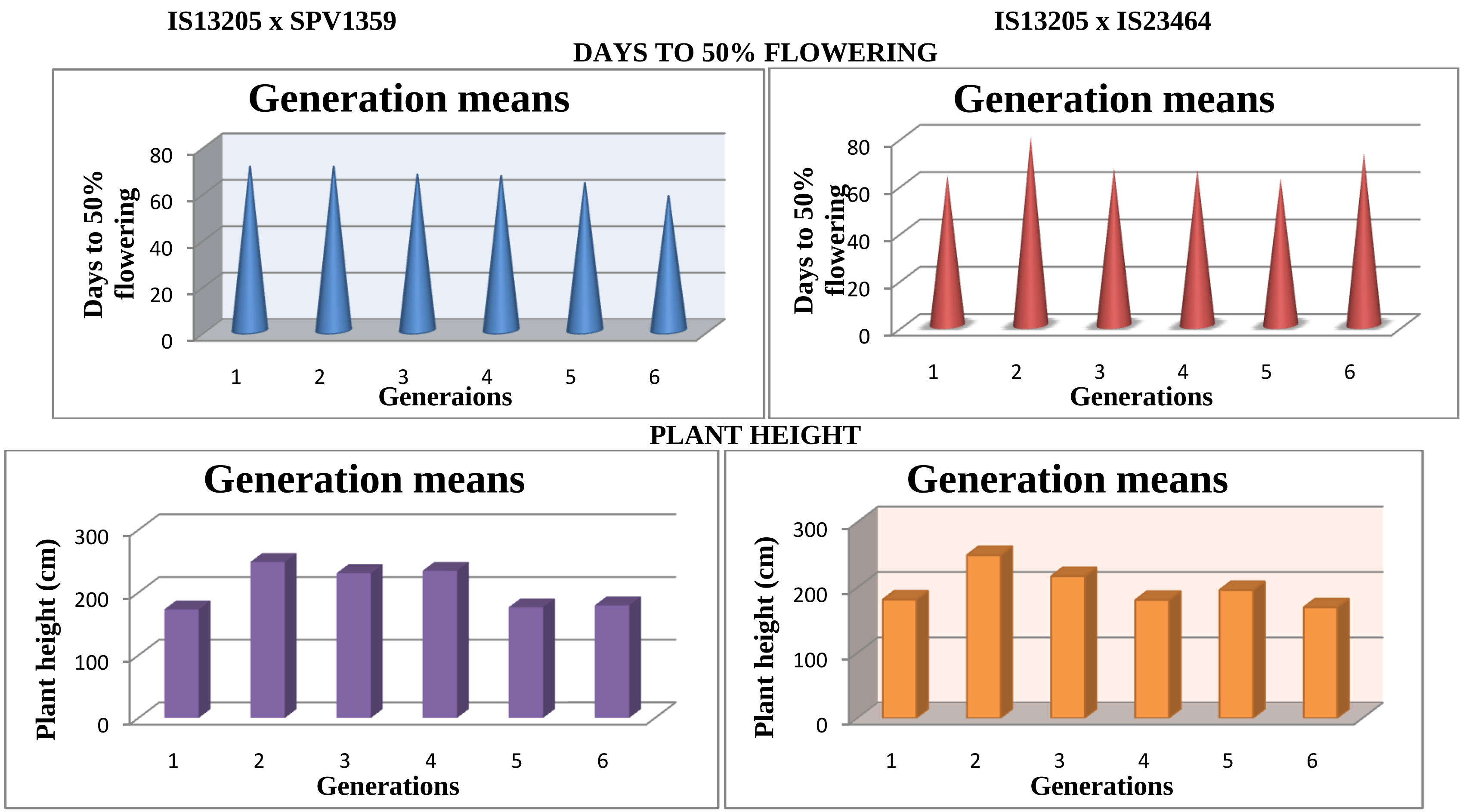


Figure 4.1. Comparison of mean performance of different generations of two crosses of sorghum for days to 50% flowering and plant height during post-rainy season, 2012-13

1- P_1, 2 - P_2, 3 - F_1, 4 - F_2, 5 - B_1, 6 - B_2

recorded slightly higher panicle length than the better parent P_1. Both B_1 and B_2 exhibited significantly longer panicles than the remaining generations and B_2 (27.9) had slightly shorter panicles than B_1 (28.82).

In the cross IS13205 × IS23464, the data from Table 4.2 revealed that parent P_1 (21.48) recorded slightly higher panicle length than the parent P_2 (19.67). The F_1 (24.09) mean was higher than the better parent. The F_2 (22.94) generation also recorded higher but panicle length than both the parents. Among the back crosses, B_1 (25.79) was almost similar to B_2 (25.52) for panicle length and both of them were significantly superior in panicle length compared than the better parent (Fig. 4.2).

4.2.4 Panicle width (cm)

Even though there was non-significant difference between the generations in the cross IS13205 × SPV1359, P_1 recorded numerically higher panicle width (11.95) than the remaining generations.

In the cross IS13205 × IS23464, panicle width exhibited non-significant difference between the generations. However, P_1 (10.82) showed slightly more panicle width. B_1 (10.02) recorded panicle width nearer to the mid parent while F_1 (8.04) and F_2 (8.08) recorded least panicle width (Fig. 4.2).

4.2.5 Grain yield plant^{-1} (g)

The data furnished in Table 4.2 revealed significant difference between the parents for grain yield plant^{-1} and parent P_2 (90.21) recorded higher yield than parent P_1 (31.16) in the cross IS13205 × SPV1359. The hybrid F_1 (36.99) recorded significantly lower yield than both mid parent and better parent. The F_2 (57.74) recorded the grain yield plant^{-1} similar to mid parent value. Back crosses B_1 (82.24) and B_2 (64.56) deviated significantly from the remaining generations for this trait.

In the cross IS13205 × IS23464, the parents deviated significantly for grain yield plant^{-1} and parent P_2 (80.09) recorded higher yield than parent P_1 (35.06). The hybrid F_1 (32.88) recorded lower grain yield plant^{-1} than mid parent value and the parent P_1, while F_2 (35.60) yielded almost on par with the parent P_1. Among the back crosses, B_2 (51.95) out yielded B_1 (34.33), which in turn yielded less than the parent P_1. Among the entire generations, parent P_2 yielded highest similar to the previous cross (Fig. 4.3).

4.2.6. Plant aspect score:

There was a significant difference between the parents for plant aspect score in the cross IS13205 × SPV1359. Parent P_1 (2.67) showed higher plant aspect score than parent P_2 (1.33). Both F_1 (2.00) and F_2 (2.00) showed same plant aspect score as that of mid parent, but it showed lesser value than the better parent. Both the back crosses B_1

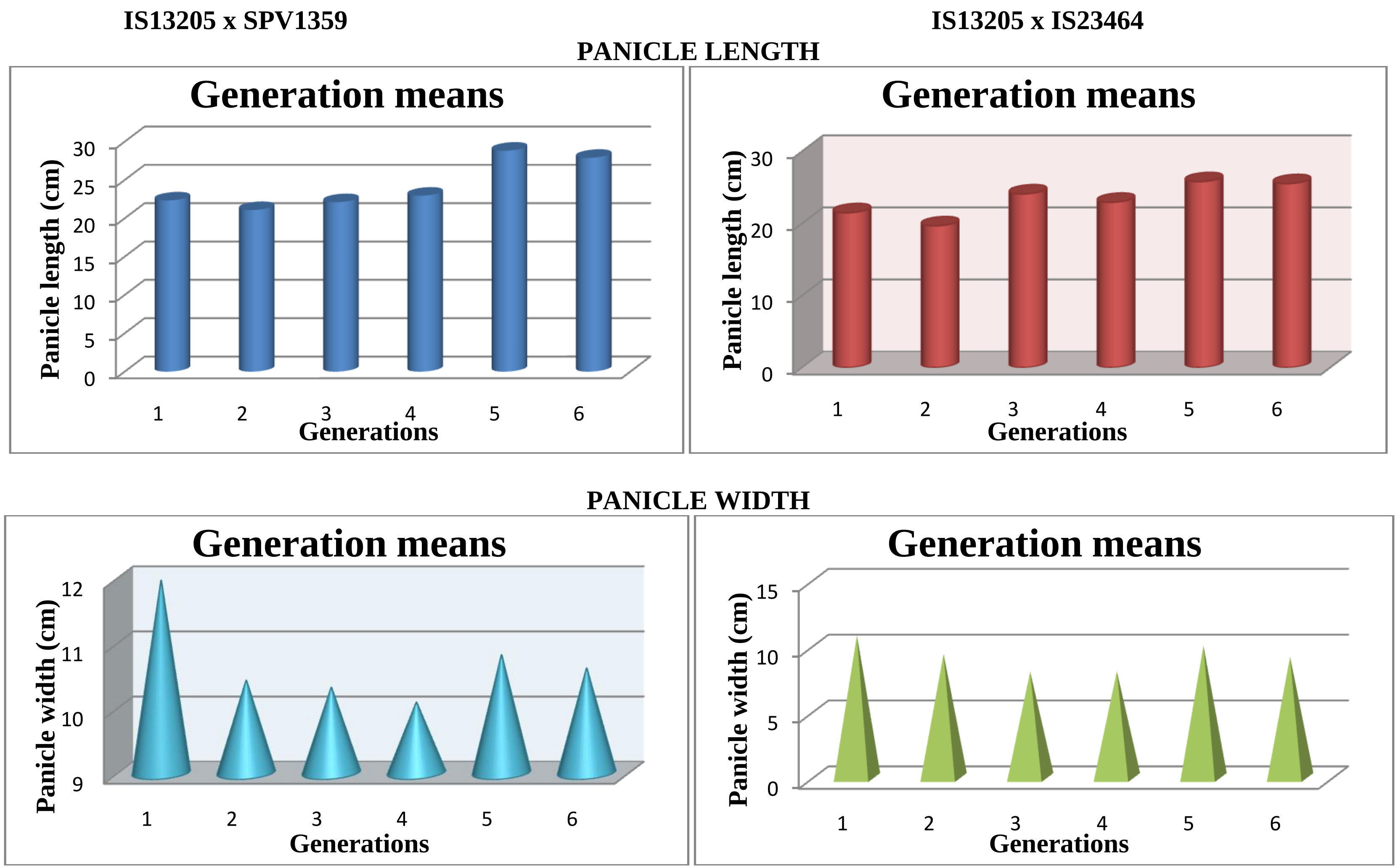


Figure 4.2. Comparison of mean performance of different generations of two crosses of sorghum for panicle length and panicle width during post-rainy season, 2012-13

1- P_1, 2 - P_2, 3 - F_1, 4 - F_2, 5 - B_1, 6 - B_2

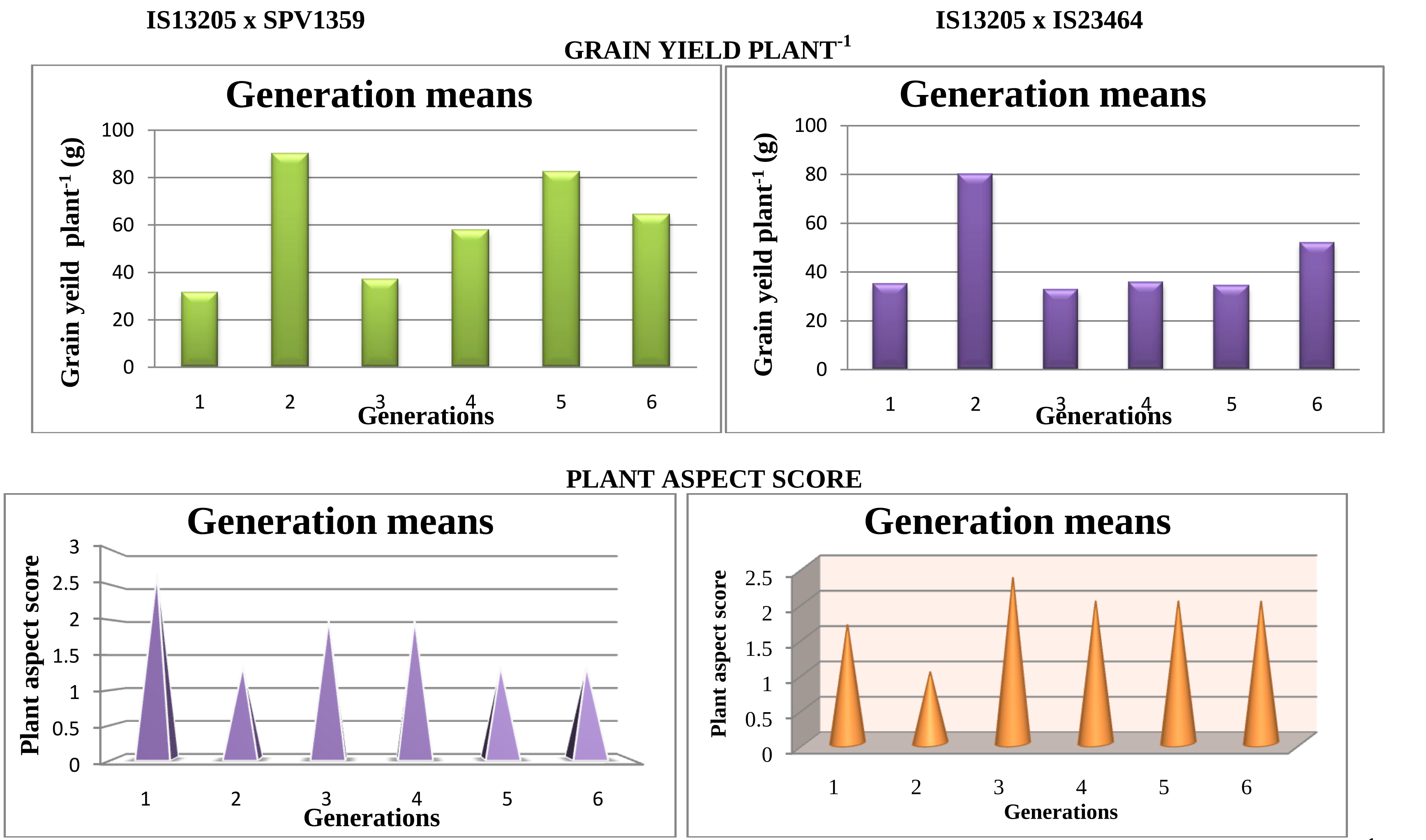


Figure 4.3. Comparison of mean performance of different generations of two crosses of sorghum for grain yield plant^{-1} and plant aspect score during post-rainy season, 2012-13

1- P_1, 2 - P_2, 3 - F_1, 4 - F_2, 5 - B_1, 6 - B_2

(1.33) and B_2 (1.33) recoded same plant aspect score. Both of them showed lesser values than mid parent and better parent for this trait.

In the cross IS13205 × IS23464, there was slight difference between the parents. Parent P_1 (1.67) recorded higher plant aspect score than parent P_2 (1.00). Both F_1 (2.33) and F_2 (2.00) out performed both mid parent and better parent. Backcrosses B_1 (2.00) and B_2 (2.00) showed same plant aspect score and also recorded higher plant aspect score than mid parent and better parent (Fig. 4.3).

4.2.7 100-grain weight (g):

Significant differences were observed between the parents for 100-grain weight and parent P_2 (4.42) recorded higher 100-grain weight than the parent P_1 (3.10) in the cross IS13205 × SPV1359 (Table 4.2). The hybrid F_1 (3.80) showed slightly higher 100-grain weight than the mid parent value, which was lesser value than that of better parent. F_2 (3.90) also showed similar results for this trait. There was no significant difference between the back crosses for 100 seed weight. But B_1 (4.31) weighed slightly higher than B_2 (4.13) and almost similar to the better parent.

In the cross IS13205 × IS23464, non-significant differences were observed among the generations for 100-grain weight. Parent P_2 recorded highest 100-grain weight (4.11), while all the remaining generations recorded almost similar results (Fig. 4.4).

4.2.8 Grain iron concentration (mg kg^{-1}):

There was significant difference between parents for grain iron concentration in the cross IS13205 × SPV1359 (Table 4.1). Parent P_1 (41.13) had higher grain iron concentration than parent P_2 (23.27). F_1 (40.37) recorded significantly higher grain iron concentration than the mid-parent but little less than better parent. F_2 (34.40) significantly deviated from F_1 for this character. Back crosses B_1 (32.13) and B_2 (29.10) differed in their grain iron concentrations while the former recorded value similar to the mid-parent, the latter recorded lesser than the mid-parent. Both the back crosses assimilated significantly less grain iron concentrations than the better parent. Between the back crosses B_1 performed better than B_2.

In the cross IS13205 × IS23464, the parents showed significant deviation for grain iron concentration (Table 4.1). Parent P_1 (39.30) recorded higher value than parent P_2 (24.43). F_1 (39.60) performed almost on a par with the better parent. F_2 (38.07) also showed similar grain iron concentration to that of better parent. Back crosses B_1 (34.50) and B_2 (35.33) showed higher grain iron concentrations than mid-parent but lesser

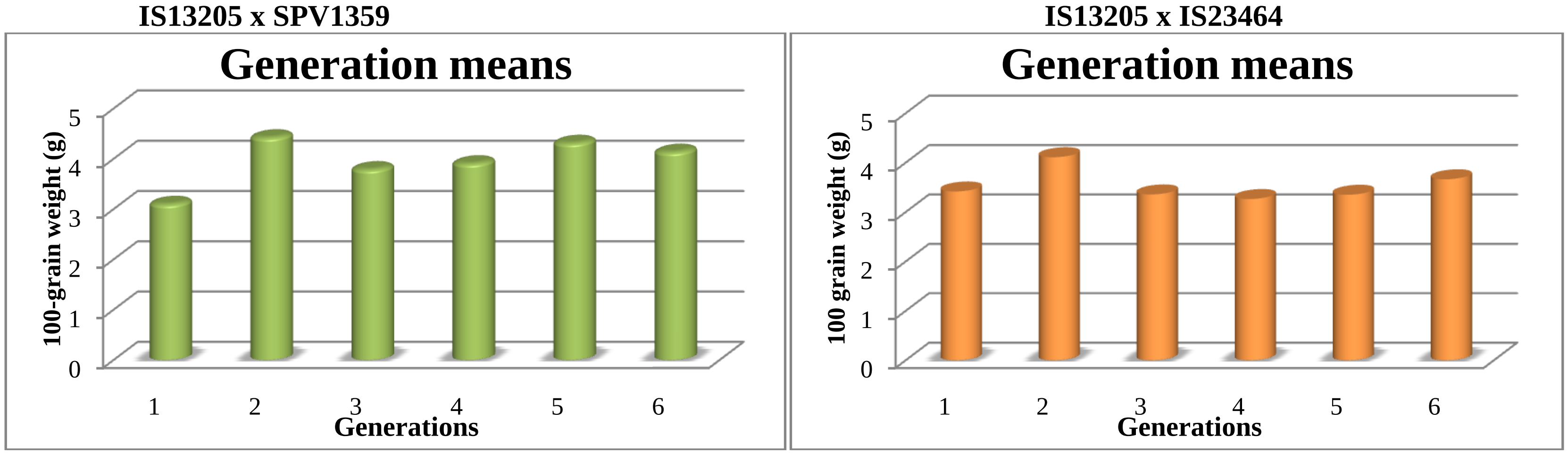
Table 4.2. Mean performance of two crosses with respect to various agronomic traits and grain iron and zinc concentrations for two crosses of sorghum during post-rainy season, 2012-13

Trait	**IS13205 x SPV1359**							
	P_1	P_2	F_1	F_2	B_1	B_2	S.Em.±	C.D. (p=0.05)
Days to 50% flowering	70.33	70.33	67.00	66.33	63.33	57.67	4.51	NS
Plant height (cm)	173.05	248.75	230.83	234.74	176.41	179.61	8.00	25.217
Panicle length (cm)	22.33	21.08	22.11	22.95	28.82	27.90	0.68	2.158
Panicle width (cm)	11.95	10.42	10.31	10.08	10.81	10.60	0.58	NS
Grain yield plant^{-1} (g)	31.16	90.21	36.99	57.74	82.24	64.56	7.45	23.469
Plant aspect score	2.67	1.33	2.00	2.00	1.33	1.33	0.29	0.919
100-grain weight (g)	3.10	4.42	3.80	3.90	4.31	4.13	0.17	0.543
Grain iron concentration (mg kg^{-1}**)**	41.13	23.27	40.37	34.40	32.13	29.10	1.54	4.863
Grain zinc concentration (mg kg^{-1}**)**	32.37	17.83	29.70	26.90	25.27	20.97	2.09	6.603

Trait	**IS13205 x IS23464**							
	P_1	P_2	F_1	F_2	B_1	B_2	S.Em.±	C.D. (p=0.05)
Days to 50% flowering	63.33	79.67	66.33	65.67	62.00	72.67	2.44	7.687
Plant height (cm)	181.10	249.18	216.88	180.47	195.78	169.28	7.04	22.168
Panicle length (cm)	21.48	19.67	24.09	22.94	25.79	25.52	0.91	2.882
Panicle width (cm)	10.82	9.40	8.04	8.08	10.02	9.19	0.67	NS
Grain yield plant^{-1} (g)	35.06	80.09	32.88	35.60	34.33	51.95	8.54	26.917
Plant aspect score	1.67	1.00	2.33	2.00	2.00	2.00	0.18	0.575
100-grain weight (g)	3.42	4.11	3.36	3.26	3.35	3.66	0.21	NS
Grain iron concentration (mg kg^{-1}**)**	39.30	24.43	39.60	38.07	34.50	35.33	1.92	6.039
Grain zinc concentration (mg kg^{-1}**)**	33.37	16.30	30.53	31.10	29.23	26.40	2.01	6.346

NS – Non Significant

100-GRAIN WEIGHT



GRAIN IRON AND ZINC CONCENTRATIONS

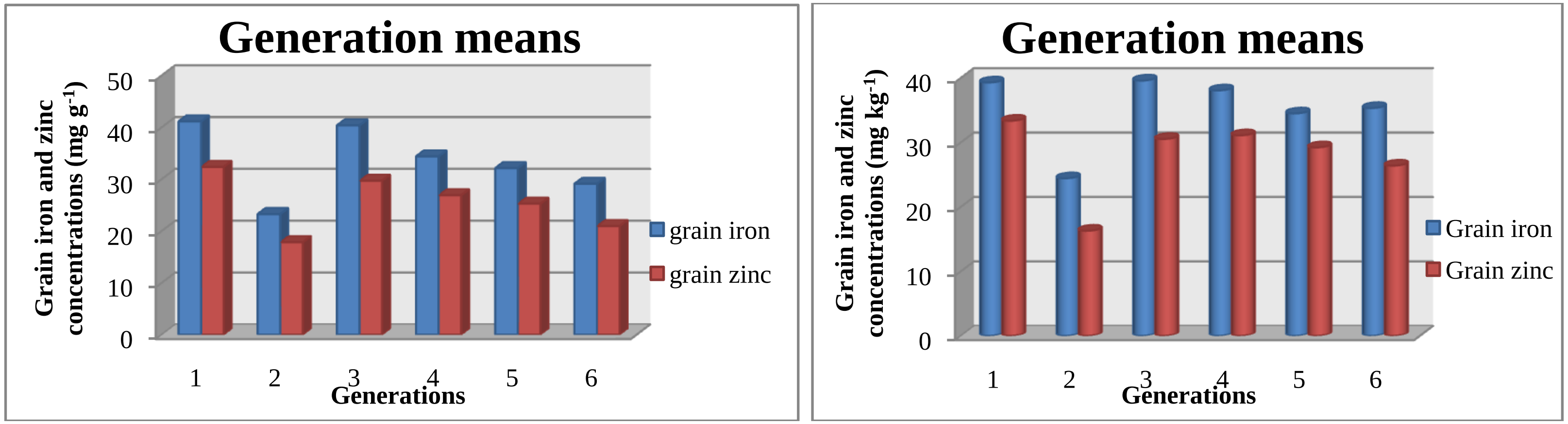


Figure 4.4. Comparison of mean performance of different generations of two crosses of sorghum for 100 grain weight and grain iron and zinc concentrations during post-rainy season, 2012-13

1- P_1, 2 - P_2, 3 - F_1, 4 - F_2, 5 - B_1, 6 - B_2

values than the better parent. Between the back crosses, B_2 performed better than B_1 (Fig. 4.4).

4.2.9 Grain zinc concentration (mg kg^{-1}):

The parents of the cross IS13205 × SPV1359 exhibited significant deviation for grain zinc concentration (Table 4.1). Parent P_1 (32.37) recorded significantly higher value than parent P_2 (17.83). F_1 (29.70) had lesser grain zinc concentration than the better parent, but had significantly higher value over mid parent. F_2 (26.90) also showed higher grain zinc concentration than that of mid parent, but less than the better parent. Among the back crosses, B_1 (25.27) showed higher grain zinc concentration than mid parent and B_2 (20.97) performed almost on par with the mid parent, but both of them showed lesser values than the better parent. Between the back crosses B_1 performed better than B_2.

In the cross IS13205 × IS23464, significant difference was observed between parents for grain zinc concentration. Parent P_1 (33.37) had significantly higher grain zinc concentration than parent P_2 (16.3). F_1 (30.53) outperformed the mid parent but performed less than the better parent. F_2 (31.1) also showed higher grain zinc concentration than the mid parent, but its value remained less than the better parent. Both the back crosses B_1 and B_2 recorded higher grain zinc concentration than the mid parent, but they assimilated lesser grain zinc concentration than the better parent. Between the back crosses, B_1 (29.23) performed better than B_2 (26.40) (Fig. 4.4).

4.3 VARIOUS GENETIC PARAMETERS:

The phenotypic coefficient of variation (PCV %), genotypic coefficient of variation (GCV %), heritability, genetic advance, genetic advance as percent of mean, degree of dominance, heterosis and inbreeding depression for eight characters (Table 4.3 and 4.4) and the results are discussed below:

4.3.1 Phenotypic Coefficient of Variation (%) and Genotypic Coefficient of Variation (%)

An assessment of heritable and non-heritable components in the total observed variability is indispensable in adopting suitable breeding procedure. The genotypic coefficient of variation (GCV%) enables the breeder to get an idea about the range of inherent variability available in the material under study so as to exploit it either through selection or hybridization. The phenotypic coefficient of variation (PCV%) gives an idea about over all variation present in the population including genotypic and

Table 4.3. Estimates of various genetic parameters for different agronomic traits and grain iron and zinc concentrations for two crosses of sorghum during post-rainy season, 2012-13

Trait	PCV %	GCV %	h^2_b	GA	GAM	h^2_n	Degree of dominance
IS13205 x SPV1359							
Days to 50% flowering	12.12	2.485	4.21	0.69	1.05	------	--------
Plant height (cm)	17.48	16.15	85.35	63.42	30.6	3.90	2.52
Panicle length (cm)	14.18	13.31	88.10	6.21	25.66	7.96	2.53
Panicle width (cm)	9.90	2.95	8.92	0.19	1.73	------	------
Grain yield plant^{-1} (g)	42.63	36.92	75.01	39.86	65.84	8.66	2.46
100-grain weight (g)	13.58	11.28	68.98	0.75	19.01	14.98	2.34
Grain iron concentration (mg kg^{-1})	21.68	19.86	86.04	12.67	37.93	5.87	2.63
Grain zinc concentration (mg kg^{-1})	24.65	19.54	65.34	8.25	32.34	7.05	2.42
IS13205 x IS23464							
Days to 50% flowering	11.02	9.12	68.45	10.53	15.42	0.90	1.38
Plant height (cm)	15.73	14.49	84.82	54.11	27.22	15.40	2.22
Panicle length (cm)	11.65	9.45	65.8	3.57	15.36	19.62	2.38
Panicle width (cm)	15.55	9.24	35.32	1.03	10.81	------	------
Grain yield plant^{-1} (g)	49.28	36.70	55.45	25.12	55.83	3.67	2.89
100-grain weight (g)	12.19	6.77	30.81	0.26	7.37	------	-------
Grain iron concentration (mg kg^{-1})	17.85	15.16	72.13	9.32	26.47	11.95	2.63
Grain zinc concentration (mg kg^{-1})	24.17	20.67	73.09	26.47	36.35	6.24	2.42

h^2_b – Heritability in broad sense

h^2_n – Heritability in narrow sense

PCV% - Phenotypic Coefficient of Variation percentage

GA – Genetic Advance

GVC% - Genotypic Coefficient of Variation percentage

GAM – Genetic Advance as percentage of Mean environmental variation from which the contribution of environment to the total variation can be known.

In the cross IS13205 × SPV1359 phenotypic coefficient of variation ranged from 9.9 for panicle width to 42.63 for grain yield plant^{-1}. Genotypic coefficient of variation varied from 2.95 for panicle width to 36.92 for grain yield plant^{-1}. In the cross, IS13205 × IS23464, PCV% ranged from 11.02 for days to 50% flowering to 49.28 for grain yield plant^{-1}. GCV% ranged from 6.77 for 100-grain weight to 36.70 for grain yield plant^{-1}.

For grain iron and zinc concentrations moderate difference between PCV% and GCV% revealed moderate influence of environment in their accumulation. In both the crosses grain zinc concentration showed higher estimates of PCV% and GCV% than iron concentration and the gap between PCV% and GCV% was higher for grain zinc concentration than grain iron concentration which implies that the accumulation of this micronutrient suffered less from environment than that of grain zinc accumulation.

The phenotypic coefficient of variation was moderately higher than genotypic coefficient of variation for all the traits under study except days to 50% flowering, which suggested moderate influence of environment on these traits. Similar results were obtained for yield and yield components by Shinde *et al*. (2010). Moderate values of PCV% and GCV% were noticed for plant height, panicle length and 100-grain weight. Higher values of PCV% and GCV% were obtained for grain yield per plant indicating that the variation for this trait contributed markedly to the total variability and high estimates of coefficient of variation for grain yield per plant was also reported earlier by Chaudhary *et al*. (2001), Sharma *et al*. (2006) and Shinde *et al*. (2010). Similar results were also reported by Warkard *et al*. (2008) and Godbharle *et al*. (2010) for all the traits studied except days to 50% flowering and panicle width, Bello *et al*. (2007) for plant height, panicle length and width and 1000-grain weight, Kumar *et al*. (2011) for all the traits, Chavan *et al*. (2010) for grain yield and Mahajan *et al*. (2011) and Seetharam and Ganeshmurthy (2013) for plant height and panicle length, Govindaraj *et al*. (2011) and Babu *et al*. (2012) for grain iron and zinc concentrations in pearlmillet and rice respectively.

4.3.2 Heritability (broad sense and narrow sense), genetic advance and degree of dominance:

Estimates of heritability (broad sense and narrow sense) and genetic advance are presented in Table 4.3. In the cross IS13205 × SPV1359, heritability in broad sense ranged from 4.21 for days to 50% flowering to 88.1 for panicle length. High heritability in broad sense was recorded for all the characters under study except days to 50%

flowering and panicle width. Heritability in narrow sense ranged from 3.90 for plant height to 14.98 for 100-grain weight. Genetic advance (GA) varied from 0.19 for panicle width to 63.42 for plant height. Genetic advance as per cent mean (GAM) ranged from 1.05 for days to 50% flowering to 65.84 for grain yield plant^{-1}. Low GAM was obtained for days to 50% flowering and panicle width. Medium GAM was observed for 100-grain weight. High GAM was recorded for panicle length, plant height, grain zinc and iron concentrations and grain yield plant^{-1}. Degree of dominance was more than one for all the traits studied which indicated overdominance.

In the cross IS13205 × IS23464, heritability in broad sense ranged from 30.81 for 100-grain weight to 84.82 for plant height. Medium heritability in broad sense was observed for 100-grain weight, panicle width and grain yield plant^{-1}while the remaining traits exhibited high heritability in broad sense. Heritability in narrow sense varied form 3.67 for grain yield plant^{-1} to 19.62 for panicle length. Genetic advance ranged from 1.03 for panicle width to 54.11 for plant height. Genetic advance as per cent mean varied form 7.37 for 100-grain weight to 55.83 for grain yield plant^{-1}. Low GAM was obtained for 100-grain weight. Medium GAM was obtained for panicle width, panicle length and days to 50% flowering. High GAM was recorded for grain iron concentration, plant height, grain zinc concentration and grain yield plant^{-1}. Degree of dominance was more than one for all the traits studied which reveals overdominance.

In both the crosses narrow sense heritability was low indicating lesser proportion of additive genetic variance in the total variance. Heritability can be determined with greater accuracy if it was studied in conjunction with genetic advance and genetic advance as percentage of mean (Johnson *et al.* 1955). Heritability values indicate only the magnitude of inheritance of the traits; whereas genetic advance is helpful in formulating the selection procedure to be adopted. High heritability (in broad sense) coupled with high genetic advance as per cent mean was observed in plant height, grain yield plant^{-1}, grain iron concentration and grain zinc concentration in both the crosses, which signifies the preponderance of additive gene action and effectiveness of selection.

Results obtained were in agreement with Godbharle *et al*. (2010), Warkard *et al*. (2008), Kumar *et al*. (2011) and Seetharam and Ganeshmurthy (2013) for plant height, panicle length and grain yield and Mahajan *et al*. (2011) for plant height and grain yield. Heritability in narrow sense ranged from 3.67 for grain yield plant^{-1} in the cross IS13205 × IS23464 to 19.62 for panicle length in cross IS13205 × IS23464. Grain zinc concentration recorded comparatively higher narrow sense heritability than the grain iron concentration. Heritability in narrow sense estimates are low for all the traits studied which signifies the lesser contribution of additive gene action.

4.4 HETEROSIS AND INBREEDING DEPRESSION:

The estimates of various heterosis parameters and inbreeding depression are presented in Table 4.4. In the cross IS13205 × SPV1359 average heterosis ranged from -4.73 for 100-grain weight to 25.37 for grain iron concentration. Significant average heterosis in positive direction was obtained for grain yield plant^{-1} and grain iron concentration revealing that the F_1 of this cross out performed mid parent significantly for grain yield plant^{-1} and grain iron concentration. Residual heterosis over mid parent ranged from -5.09 for grain yield plant^{-1} to 11.18 for plant height. Significant residual heterosis over mid parent was obtained for plant height, though average heterosis was not significant which might be due to the fixation of alleles in F_2. Heterobeltiosis ranged from -0.98 for panicle length to -58.99 for grain yield plant^{-1}. Significant negative heterobeltiosis was obtained for grain yield plant^{-1} and 100-grain weight. All the characters showed negative estimates of heterobeltiosis irrespective of significance which implied that the F_1 performance was towards the poorer parent for all the traits which was particularly desirable for plant height and days to 50% flowering. Residual heterosis over better parent ranged from -5.63 for plant height to 2.78 for panicle length. Residual heterosis over better parent was negatively significant for panicle width, grain yield plant^{-1} and grain iron concentration. Thus only the F_2 population retained some heterosis for only grain yield plant^{-1}. Inbreeding depression ranged from -1.00 for days to 50% flowering to 56.07 for grain yield plant^{-1}. Significant inbreeding depression was obtained for grain iron concentration. In the cross IS13205 × IS23464, average heterosis ranged from -7.23 for 100-grain weight to 24.39 for grain iron concentration. Significant average heterosis was obtained in positive direction for grain iron and zinc concentrations and in negative direction for panicle length and width. Residual heterosis over mid parent ranged from -8.15 for days to 50% flowering to 24.5 for grain zinc concentration. Significant residual heterosis over mid parent was obtained in positive direction for grain iron and zinc concentrations and in negative direction for plant height and panicle width. Thus F_2 population could retain some heterosis for panicle width, grain iron and zinc concentrations. Heterobeltiosis varied from -8.51 for grain zinc concentration to 12.15 for panicle length. Significant negative heterobeltiosis was recorded for days to 50% flowering, plant height, panicle width, grain yield plant^{-1} and 100-grain weight.

Table 4.4. Estimates of heterosis parameters and inbreeding depression for various agronomic traits and grain iron and zinc concentrations for two crosses of sorghum during post-rainy season, 2012-13

Trait	Average Heterosis	RHM	Heterobeltiosis	RHB	ID
IS13205 x SPV1359					
Days to 50% flowering	9.45	-5.69	-4.73	-5.69	-1.00
Plant height (cm)	1.85	11.18*	-7.20	-5.63	1.69
Panicle length (cm)	-7.83	5.73	-0.98	2.78	3.81
Panicle width (cm)	-39.18	-9.86	-13.72	-15.64*	-2.20
Grain yield plant^{-1} (g)	0.79*	-5.09	-58.99**	-35.99*	56.07
100-grain weight (g)	-4.73	3.91	-14.25*	-11.67	3.08
Grain iron concentration (mg kg^{-1})	25.37**	6.83	-1.85	-16.76**	14.78*
Grain zinc concentration (mg kg^{-1})	18.32	7.17	-2.67	-16.89	9.43
IS13205 x IS23464					
Days to 50% flowering	0.81	-8.15	-16.74**	-17.51**	-0.99
Plant height (cm)	17.13	-16.12**	-12.96**	-27.00**	-16.79**
Panicle length (cm)	-20.42*	11.51	12.15	6.77	-4.79
Panicle width (cm)	-42.89*	-20.06*	-25.69*	-25.32*	0.46
Grain yield plant^{-1} (g)	-10.9*	-38.18	-58.95**	-55.55**	8.25
100-grain weight (g)	-7.23	-13.22	-18.25*	-20.61*	-2.80
Grain iron concentration (mg kg^{-1})	24.39**	6.21*	0.76	-3.13	3.86
Grain zinc concentration (mg kg^{-1})	22.22*	24.5*	-8.51	-6.80	-1.86

*****– Significant at 5% level of probability **– Significant at 1% level of probability

RHM – Residual Heterosis over Mid parent

RBH – Residual Heterosis over Bette Parent

ID – Inbreeding Depression

Heterobeltiosis estimates are negative for most of the traits as in the previous cross irrespective of the significance which indicates that the performance of F_1 was towards poor parent. Residual heterosis over better parent ranged from -3.13 for grain iron concentration to 6.77 for panicle length. Significant residual heterosis over better parent was obtained for days to 50% flowering, plant height, panicle width, grain yield plant^{-1} and 100-grain weight in negative direction. Thus, the F_2 population could retain some heterosis over better parent for the traits which showed significant heterobeltiosis. Inbreeding depression ranged from -0.99 for days to 50% flowering to 8.25 for grain yield plant^{-1}. Significant negative inbreeding depression was obtained for plant height which might be desirable as dwarf plants are resistant to lodging.

Significant heterosis (either average heterosis or heterobeltiosis) coupled with significant inbreeding depression signifies non-additive gene action where as significant heterosis coupled with non-significant inbreeding depression implies additive gene action. For most of the traits studied in both the crosses, the second situation was observed, which reveals the importance of additive gene action in their inheritance. However, it is highly unrealistic to come to a conclusion based on two parameters and in only two crosses. Hence further study is need for making valid conclusions.

Significant positive average heterosis for grain yield was also obtained by Choudhari (1992), Rao *et al*. (1993), Shankarapandian *et al*. (1994) and Ganesh *et al.* (1996). Significant average heterosis for grain iron and zinc concentrations were also recorded by Velu *et al*. (2011) and Govindraj *et al*. (2013) in pearl millet and Ashok Kumar *et al*. (2013) in sorghum.

Significant negative heterosis over better parent was reported for days to 50% flowering, plant height by Pawar (2000) and Lokapur (1997) and for 100 seed weight by Desai *et al*. (1985). Results obtained for grain iron and zinc concentrations for heterobeltiosis were in accordance with Velu *et al*. (2011), Govindaraj *et al*. (2013) in pearl millet and Gayathri (2012) and Ashok Kumar *et al*. (2013) in sorghum.

4.5 CORRELATION STUDIES:

The study of relationships among quantitative traits is important for assessing the feasibility of joint selection of two or more traits and hence evaluating the effect of selection for secondary traits in genetic gain for the primary trait under consideration. A positive genetic correlation between two desirable traits makes the job of the plant breeder easy for improving both traits simultaneously. Even the lack of correlation is useful for the joint improvement of the two traits. Clear insight on genetic associations of the grain iron and zinc concentrations with grain yield and its component traits can help breeders in devising a suitable breeding strategy for enhancement of micronutrient density in sorghum. Hence in the present study, the segregating population *i.e.*, F_2 of two crosses was utilized for calculation of correlation coefficients between the traits for each cross separately. The results of the correlation analysis were presented in the Table 4.5.

In the cross IS13205 × SPV1359, panicle length showed significant positive correlation with panicle width (r = 0.651), grain yield plant^{-1} (r = 0.425) and grain zinc concentration (r = 0.328). However, the correlation was strong with panicle width and weaker with grain zinc concentration as indicated by the correlation coefficient estimates. Grain iron concentration showed highly significant positive correlation with grain zinc concentration (r = 0.538).

In the cross IS13205 × IS23464, grain yield plant^{-1} showed significant negative correlation with 100-grain weight (r = -0.390) whose magnitude was less. As in the previous cross, grain iron concentration showed highly significant positive correlation with grain zinc concentration (r = 0.753). Similar results of significant positive correlation coefficient between iron and zinc was also obtained by Graham and Welch (1996) in general, Reddy *et al*. (2005), Ashok Kumar *et al*. (2009, 2010) and Gayathri *et al*. (2012) in sorghum, Nagesh *et al*. (2012) in rice, Morgunove *et al*. (2007), Zhao *et al*. (2009), Yong *et al*. (2010), Xian *et al*. (2010) and Feng *et al*. (2011) in wheat, Velu *et al*. (2008, 2011), Govindaraj *et al*. (2009, 2013) and Gupta *et al*. (2009) in pearl millet and Oikeh *et al*. (2003) and Chakraborthi *et al*. (2011) in maize. Non-correlation of grain iron and zinc concentrations with grain yield plant^{-1} was also observed by Nagesh *et al*. (2012) in rice, Chakraborthi *et al*. (2011) in maize and Gupta *et al*. (2009) in pearl millet. No correlation of test weight with grain iron and zinc concentrations was also recorded by Gupta *et al*. (2009) in pearl millet and Wang *et al*. (2011) in wheat.

Highly significant positive correlation between grain iron and zinc concentrations indicated the possibility of simultaneous improvement of both the traits. This might be due to co-segregation of tightly linked genetic elements governing the physiology of these micronutrients or might be due to the pleotropic effect of genes. No correlation of 100-grain weight and grain yield plant^{-1} with grain iron and zinc concentrations indicate that there would be no penalty on grain yield and seed size while breeding for grains rich in these micronutrients. However, negative correlation was reported by many authors between grain yield plant^{-1} and grain iron and zinc concentrations. Moreover, grain yield plant^{-1} showed negative correlation with 100-

Table 4.5. Simple correlation matrix of grain iron and zinc concentrations with other agronomic traits in sorghum during post-rainy season, 2012-13

	Plant height (cm)	**Panicle length (cm)**	**Panicle width (cm)**	**Grain yield plant^{-1} (g)**	**100-grain weight (g)**	**Grain iron concentration (mg kg^{-1})**	**Grain zinc concentration (mg kg^{-1})**
IS13205 x SPV1359							
Plant height (cm)	**1.000**	0.116	0.008	0.172	0.197	-0.227	-0.023
Panicle length (cm)		**1.000**	0.651**	0.425**	-0.006	0.094	0.328*
Panicle width (cm)			**1.000**	0.135	-0.025	-0.105	0.062
Grain yield plant^{-1} (g)				**1.000**	0.142	0.181	0.127
100-grain weight (g)					**1.000**	-0.053	-0.199
Grain iron concentration (mg kg^{-1})						**1.000**	0.538**
Grain zinc concentration (mg kg^{-1})							**1.000**
IS13205 x IS23464							
Plant height (cm)	**1.000**	0.179	0.024	0.179	0.042	0.267	0.266
Panicle length (cm)		**1.000**	0.242	0.093	0.005	0.007	0.008
Panicle width (cm)			**1.000**	0.161	-0.102	0.099	0.057
Grain yield plant^{-1} (g)				**1.000**	-0.390*	0.083	0.002
100-grain weight (g)					**1.000**	0.133	0.165
Grain iron concentration (mg kg^{-1})						**1.000**	0.753**
Grain zinc concentration (mg kg^{-1})							**1.000**

* – Significant at 5% level of probability *i.e.*, r = 0.31198

** – Significant at 1% level of probability *i.e.*, r = 0.40262 grain weight which might be due to indirect effects via other possible traits which were not included in the study. Hence, there is a need to confirm the results of present study by using various types of populations like inbreds, hybrids and OPVs.

The estimates of correlations alone may be often misleading due to mutual cancellation of component traits (Mahajan *et al*., 2011). So it becomes necessary to do path coefficient analysis which takes into account the causal relationship in addition to the degree of relationship. However, it can be seen from the correlation matrix in Table 4.5 that the estimates of correlation coefficients obtained in the present investigation were very low among the traits with significant correlations obtained in only few cases. Hence, the data were not subjected to path coefficient analysis.

4.6 GENE EFFECTS:

Estimation of the types of gene action involved in the expression of traits, the level of additive effects and the degree of dominance are very important in designing a breeding method for improving the trait of interest. Knowledge of the way genes act and interact will determine which breeding system can optimize gene action more efficiently and will help to elucidate the role of breeding systems in the evolution of crop plants (Hallauer and Miranda, 1988). The information on genetics of grain iron and zinc concentrations in sorghum is limited. Therefore, in the present investigation, an attempt was made to understand the genetics of grain iron and zinc concentrations along with some important agronomic traits through generation mean analysis, a biometrical method developed by Mather and Jinks (1982), which greatly helps in the estimation of various components of genetic variance.

The mean data obtained from six generations of the two cross combinations for yield, yield components, grain iron and zinc concentrations were subjected to generation mean analysis using scaling testes to test the fitness of additive-dominance model and Hayman’s six parameter model to find the significant inter-allelic interactions. The data on gene effects are presented in Table 4.6 and 4.7 and described only for those traits where mean sum of squares due to generations were found significant.

4.6.1 Days to 50% flowering:

The three scaling test *viz,*. A, B and C were non-significant for this trait in the cross IS13205 × IS23464 for which there was significant differences among generations. This indicates the adequacy of simple additive dominance model to explain the inheritance of this character and the absence of interactions. Using three parameter model (Mather, 1949), the additive and dominance variances were calculated and the estimates revealed the predominance of dominance component over additive component. However as this conclusion was based on a single cross, it was unreliable and a more extensive study using more number of crosses will be necessary in future for further confirmation. Absence of epistasis for days to flowering was also observed by Raju (1979) and Desai and Shukla (1995) in summer season in sorghum.

4.6.2 Plant height (cm):

The three scaling tests *viz*., A, B and C were significant in the cross IS13205 × SPV1359, while B and C were significant in the cross IS13205 × IS23464, indicating the inadequacy of simple additive-dominance model and the presence of epistatic interactions. Hence a six parameter model was adopted to test the presence of non-allelic interactions.

The partitioning of generation means and estimation of genetic components revealed highly significant positive mean value in both the crosses studied. In the cross IS13205 × SPV1359, all the genetic parameters were significant. The estimates of additive, dominance and additive × additive were negatively significant while the additive × dominance and dominance × dominance estimates were positively significant. The value of dominance component was more compared to additive component. Among interactions additive × additive and dominance × dominance were of higher values. The results obtained were in line with those of Indi and Goud (1981) and Desai and Shukla (1995) in sorghum.

In the cross IS13205 × IS23464, only additive component was significant in positive direction where as the dominance component was non-significant. Among interactions, only additive × dominance was significant in positive direction while additive × additive and dominance × dominance were non-significant. With regard to magnitude, only significant genetic components were higher in magnitude. The results from both the crosses were quite contradictory both in direction and significance except the component additive × dominance. Hence there is a need for a detailed study of this character using more number of crosses involving contrasting parents.

4.6.3 Panicle length (cm):

Three scaling tests, *viz*., A, B and C were significant in the cross IS13205 × SPV1359 and A and B were significant in the cross IS13205 × IS23464 which indicates the inadequacy of simple additive dominance model in explaining the gene action involved in the inheritance of this character.

The partitioning of generation means into six components revealed that the mean component (*m*) was positive and highly significant in both the crosses under study. In both the crosses, only dominance component was significant in positive direction while the additive component was non-significant. Among the interactions, additive × additive and dominance × dominance were significant in positive and negative directions, respectively. With regard to the magnitude, dominant component was of much higher value compared to additive component where as dominance × dominance was of higher value followed by additive × additive, though the former was negative and the latter was positive. However, the magnitudes of various components were lesser in the cross IS13205 × IS23464 compared to the cross IS13205 × SPV1359. The results in both the crosses are similar for panicle length which proved the reliability of the results obtained. Similar results were obtained by Indi and Goud (1981), Biradar and Borikar (1985), Desai and Shukla (1995) and Narain *et al*. (2007) in sorghum.

4.6.4 Grain yield plant^{-1} (g):

Only one scaling test i.e., A was significant in the cross IS13205 × SPV1359 and scaling test C was significant in the cross IS13205 × IS23464, which indicated the failure of additive-dominance model to explain the inheritance of grain yield plant^{-1}.

The partitioning of generation means into six components revealed that the component (*m*) was positive and highly significant in both the crosses used in the present investigation. In the cross IS13205 × SPV1359, additive and dominance components were significant in positive direction. Among interactions, additive × additive and additive × dominance were significant in positive direction where as dominance × dominance was significant in negative direction. With regard to the magnitude, dominance component was of higher value compared to additive and among interactions dominance × dominance was higher in magnitude but in negative direction. The results obtained were in line with those of Nayakar (1973), Indi and Goud (1981), Nimbalkar *et al*. (1988), Rao *et al*. (1994), Desai and Shukla (1995) and Rani *et al*. (2008) in sorghum.

In the cross IS13205 × IS23464, additive component was negatively significant while the dominance component was non-significant. Among the interactions additive × additive was significant in positive direction where as the remaining interactions, i.e., additive × dominance and dominance × dominance were non-significant. The value of additive component was higher compared to dominance component where as among interactions additive × additive interaction was of highest value. The results of this cross were similar to those obtained by Narain *et al*. (2007). These results were completely different from the results of previous cross as in the case of plant height. Hence there is an absolute need to probe the inheritance of this character using more number of crosses to enhance the authenticity of the conclusions.

4.6.5 100-grain weight (g):

Only one scaling test i.e., A was significant in the cross IS13205 × SPV1359 even then it indicated the presence of non-allelic interactions and inadequacy of additive-dominance model in explaining the inheritance of this character in this cross. Since this trait exhibited non-significant deviation among the generations of the cross, IS13205 × IS23464, generation mean analysis was not carried out.

The partitioning of generation means into six components revealed that the mean component (*m*) was positive and highly significant in the cross IS13205 × SPV1359, for which significant differences among generations were obtained. Both additive and dominance components were insignificant whereas, among interactions, only additive × dominance was significant in positive direction. Remaining interactions i.e., additive × additive and dominance × dominance were insignificant. With regard to the magnitude both additive and dominance components were similar, though non-significant whereas among interactions additive × dominance was of highest magnitude. This unclear situation may be due to the presence of higher order interactions or linkage effects which needs further analysis with enough generations to fit a full trigenic interaction and linkage model. Significance of additive × dominance for this trait was also obtained by Nimbalkar *et al*. (1986) in sorghum.

4.6.6 Grain iron concentration (mg kg^{-1}):

Only one scaling test *i.e*., A in the cross IS13205 × SPV1359 and C in the cross IS13205 × IS23464, was significant indicating the inadequacy of additive-dominance model to explain the inheritance of grain iron concentration.

The partitioning of generation means into six components revealed that the component (*m*) was positive and highly significant in both the crosses. In the cross IS13205 × SPV1359, neither the additive component nor the dominance component was significant which indicated lesser importance of direct effects. All the inter-allelic interactions are highly significant with additive × additive and additive × dominance significant in negative direction and dominance × dominance in positive direction. With regard to magnitude dominance component was higher in magnitude than the additive component. Among interactions, dominance × dominance was of highest magnitude followed by additive × additive and additive × dominance.

Table 4.6. Results of scaling tests and genetic components for various agronomic traits and grain iron and zinc concentrations in the cross IS13205 x SPV1359 during post-rainy season, 2012-13

Scaling test	Plant height (cm)	Panicle length (cm)	Grain yield plant^{-1} (g)	100-grain weight (g)	Grain iron (mg kg^{-1})	Grain zinc (mg kg^{-1})
A	-54.33±13.09**	13.5±1.93**	95.985±11.64**	1.412±0.23**	-17.234±3.08**	-11.53±3.21**
B	-116.91±14.32**	12.568±1.35**	0.119±13.83	-0.46±0.42	-5.434±2.86	-5.59±1.88**
C	62.4±17.59**	5.002±2.02**	-1.134±14.34	0.384±0.34	-7.414±4.14	-2±2.92

Genetic component	Plant height (cm)	Panicle length (cm)	Grain yield plant^{-1} (g)	100-grain weight (g)	Grain iron (mg kg^{-1})	Grain zinc (mg kg^{-1})
m	235.81±1.94**	23.16±0.32**	48.75±2.33**	3.893±0.05**	34.43±0.76**	26.9±0.53**
d	-7.46±7.15**	1.13±0.96	18.51±7.59**	0.276±0.21	3.03±1.73	4.3±1.63*
h	-213.22±18.09**	21.39±2.45**	73.64±18.7**	0.648±0.48	-7.08±4.82	-10.52±4.03*
i	-233.64±16.28**	21.06±2.32**	97.24±17.83**	0.568±0.46	-15.25±4.62**	-15.12±3.91**
j	31.29±8.64**	0.46±1.11	47.93±8.6**	0.936±0.23**	-5.9±1.97**	-2.97±1.79
l	404.88±33.6**	-47.13±4.37**	-193.34±33.72**	-1.52±0.9	37.92±8.08**	32.24±7.17**

*****– Significant at 5% level of probability

**– Significant at 1% level of probability

m – mean   *i* – additive x additive

d – additive   *j* – additive x dominance

h – dominance   *l* – dominance x dominance

Table 4.7. Results of scaling tests and genetic components for various agronomic traits and grain iron and zinc concentrations in the cross IS13205 x IS23464 during post-rainy season, 2012-13

Scaling test	Days to 50% flowering	Plant height (cm)	Panicle length (cm)	Grain yield plant^{-1} (g)	Grain iron (mg kg^{-1})	Grain zinc (mg kg^{-1})
A	7.59±9.67	0.7±17.7	7.87±2.12**	4.433±8.07	-5.83±3.99	-5.11±2.3*
B	6.59±6.13	-118.327±20.46**	6.49±2.04**	-14.25±13.59	4.97±3.49	6.44±2.84*
C	0.52±8.29	-147.74±15.98**	2.96±2.46	-45.929±15.73**	14.94±5.03**	13.95±2.71**

Genetic component	Days to 50% flowering	Plant height (cm)	Panicle length (cm)	Grain yield plant^{-1} (g)	Grain iron (mg kg^{-1})	Grain zinc (mg kg^{-1})
m	NI	178.6067±2.56**	22.95±0.28**	35.32±1.2**	38±0.79**	30.97±0.53**
d	NI	26.93±12.27*	0.94±1.2	-16.29±4.24**	0.9±2.17	2.83±1.68
h	NI	34.0297±27.31	14.32±2.87**	7.5195±12.3	-10.99±5.73	-7.185±4.06
i	NI	30.1132±26.61	11.4±2.65**	36.112±9.76**	-15.8±5.38**	-12.62±3.98**
j	NI	59.5135±13.34**	0.69±1.3	9.3415±7.12	-5.4±2.36*	-5.775±1.78**
l	NI	87.5138±51.63	-25.76±5.4**	-26.295±23.15	16.66±10.05	11.29±7.24

***** – Significant at 5% level of probability

** – Significant at 1% level of probability

NI – No Interaction

m – mean | *i* – additive x additive

d – additive | *j* – additive x dominance

h – dominance | *l* – dominance x dominance

In the cross IS13205 × IS23464, both additive and dominance gene actions were non-significant as in the previous cross. Among interactions, additive × additive and additive × dominance were significant in negative direction, but dominance × dominance was non-significant in contrast to the previous cross. Dominance component was of higher magnitude but in negative direction compared to additive component. Among interactions, dominance × dominance was of highest magnitude, though not significant followed by additive × additive and additive × dominance interactions. The component dominance × dominance was significant in only in one cross which might due to the difference in the material used as parents.

4.6.7 Grain zinc concentration (mg kg^{-1}):

Two scaling test, A and B in the cross IS13205 × SPV1359 and all the three scaling tests in the cross IS13205 × IS23464 were significant showing the inadequacy of additive-dominance model to explain the inheritance of grain zinc concentration.

The partitioning of generation mean and estimation of genetic components revealed highly significant positive mean value in both the crosses studied. In the cross IS13205 × SPV1359, both additive and dominance components were significant though former was significant in positive direction while the latter was significant in negative direction. Among interactions additive × additive was significant in negative direction where as dominance × dominance was significant in positive direction. Additive × dominance was non-significant. With regard to the magnitude, dominance was of higher value as in the case of grain iron concentration. Among interactions, dominance × dominance was of highest value followed by additive × additive and additive × dominance.

In the cross IS13205 × IS23464, both additive and dominance components were non-significant in contrast to the previous cross. Among the interactions, additive × additive and additive × dominance were significant in negative direction where as dominance × dominance was non-significant. With regard to the magnitude, dominance component was of higher value in negative direction compared to additive component. Among interactions, dominance × dominance interaction was the highest followed by additive × additive and additive × dominance. The results of two crosses were quite different except in case of additive × additive interactions; hence the reliability of results obtained in the present investigation needs to be checked by further studies using more number of crosses.

4.6.8 Comparison of gene effects over traits in two crosses:

The comparison of gene effects over traits in two crosses was presented in the Table 4.8. In the cross IS13205 × SPV1359, mean value were highly significant for all the traits studied. Additive component was significant for plant height (in negative direction), grain yield plant^{-1} and grain zinc (in positive direction) and it was non-significant for panicle length and grain iron concentration. The dominance component was significant for the traits plant height and grain zinc concentration in negative direction and panicle length and grain yield plant^{-1} in positive direction whereas for 100-grain weight and grain iron concentration it was non-significant. Among the interactions, additive × additive interaction was significant for almost all the traits studied except 100-grain weight and for plant height, grain iron and zinc concentrations its significant value was in negative direction. Additive × dominance interaction was significant for all the traits studied except panicle length and grain zinc concentration. Its significant value was in negative direction for grain iron concentration only. Dominance × dominance interaction was significant for all the traits studied, except 100-grain weight, whose significant values were in negative direction for panicle length and grain yield plant^{-1} and in positive direction for remaining traits.

In the cross IS13205 × IS23464, mean values were highly significant for all the traits studied. Additive component was significant for plant height and grain yield per plant in positive and negative direction, respectively while it was non-significant for panicle length, grain iron and zinc concentrations. Dominance component was positively significant for panicle length, but non-significant for plant height, grain yield plant^{-1}and grain iron and zinc concentrations. Among interactions additive × additive was significant for panicle length and grain yield plant^{-1} in positive direction and for grain iron and zinc concentrations in negative direction. Additive × dominance was positively significant for plant height, but it was negatively significant for grain iron and zinc concentrations and dominance × dominance interaction was significant only in panicle length in negative direction. The differences between the generations were non-significant for 100-grain weight in this cross and hence it was not subjected to generation mean analysis.

Comparison among the gene effects revealed that dominance component was higher in magnitude than the additive effects for all the traits studied except for grain yield plant^{-1} in the cross IS13205 × IS23464. Additive × additive interaction had higher value than additive component and dominance × dominance interaction was of higher magnitude than the corresponding dominance component for all the traits studied,

Table 4.8. Comparison of gene actions for various traits in two crosses in sorghum during post-rainy season, 2012-13

Trait	*m*	*d*	*h*	*i*	*j*	*l*	Epistasis
IS13205 x SPV1359							
Plant height (cm)	235.81±1.94**	-7.46±7.15**	-213.22±18.09**	233.64±16.28**	31.29±8.64**	404.88±33.6**	Duplicate
Panicle length (cm)	23.16±0.32**	1.13±0.96	21.39±2.45**	21.06±2.32**	0.46±1.11	-47.13±4.37**	Duplicate
Grain yield plant^{-1} (g)	48.75±2.33**	18.51±7.59**	73.64±18.7**	97.24±17.83**	47.93±8.6**	-193.34±33.72**	Duplicate
100-grain weight (g)	3.893±0.05**	0.276±0.21	0.648±0.48	0.568±0.46	0.936±0.23**	-1.52±0.9	Duplicate
Grain iron (mg kg^{-1})	34.43±0.76**	3.03±1.73	-7.08±4.82	-15.25±4.62**	-5.9±1.97**	37.92±8.08**	Duplicate
Grain zinc (mg kg^{-1})	26.9±0.53**	4.3±1.63*	-10.52±4.03*	-15.12±3.91**	-2.97±1.79	32.24±7.17**	Duplicate
IS13205 x IS23464							
Days to 50% flowering	NI	NI	NI	NI	NI	NI	
Plant height (cm)	178.6067±2.56**	26.93±12.27*	34.0297±27.31	30.1132±26.61	59.5135±13.34**	87.5138±51.63	Complementary
Panicle length (cm)	22.95±0.28**	0.94±1.2	14.32±2.87**	11.4±2.65**	0.69±1.3	-25.76±5.4**	Duplicate
Grain yield plant^{-1} (g)	35.32±1.2**	-16.29±4.24**	7.5195±12.3	36.112±9.76**	9.3415±7.12	-26.295±23.15	Duplicate
Grain iron (mg kg^{-1})	38±0.79**	0.9±2.17	-10.99±5.73	-15.8±5.38**	-5.4±2.36*	16.66±10.05	Duplicate
Grain zinc (mg kg^{-1})	30.97±0.53**	2.83±1.68	-7.185±4.06	-12.62±3.98**	-5.775±1.78**	11.29±7.24	Duplicate

***** – Significant at 5% level of probability

** – Significant at 1% level of probability

NI – No Interaction

m – mean　　*i* – additive x additive

d – additive　　*j* – additive x dominance

h – dominance　　*l* – dominance x dominance which indicated the involvement of epistatic interactions. Dominance × dominance had higher value compared to additive × additive interaction which indicated the predominance of dominance gene action in the expression of the traits. In contrast Chakraborthi *et al*. (2010) observed the predominance of additive gene action for kernel iron and dominance gene action for kernel zinc where as among interactions additive × dominance was of higher magnitude for kernel iron and additive × additive was of higher magnitude for kernel zinc in maize. Gayathri (2012) and Ashok Kumar *et al*. (2013) reported the importance of non-additive gene action for grain iron concentration and additive gene action for grain zinc concentration in sorghum and Velu *et al*. (2011) and Govindraj *et al*. (2013) highlighted the importance of additive gene action in the inheritance of grain iron and zinc concentrations in pearl millet through combining ability analysis. Thus gene actions for grain/kernel iron and zinc concentrations were different in different crops. Further within the crop, they may vary with the kind of material and the diversity of the parents used, the sampling procedure followed and the associated interactions like linkage and epistasis.

The positive values of dominance (*h*) observed in most cases except for grain iron and zinc concentrations in both the crosses, indicated that the alleles responsible for high value of the trait were dominant over the alleles controlling low value whereas reverse was true for grain iron and zinc concentrations. In present study, both additive and dominance components were significant for most of the traits studied. The results also indicated the important role of digenic non-allelic interactions (epistasis), and therefore point out more complex mechanisms of genetic control. But with the predominance of dominance component over additive component and the importance of epistatic interactions, hybrid breeding can be a better strategy for sorghum improvement provided hybrid seed production is relatively simple and economically viable. Recurrent selection, biparental mating and diallel selective mating system may also be profitable to exploit both additive and non-additive components for bringing about improvement in grain yield and its attributes. Such a strategy will help in increasing the frequency of favorable alleles while maintaining the genetic variation in breeding population (Doerksen *et al*., 2003). High estimates of dominance effects compared to the additive effects are due to heterozygosity of those genes for which the parents are differing (Kearsy and Pooni, 1996) and also due to high degree of dispersion of increasing alleles between the parents. However, sometimes additive genetic effects are being underestimated because of the lack of knowledge about parental differences regarding investigated traits (Wilson *et al*., 2000). The same authors further emphasized that, for reliable estimates of genetic effects using generation mean analysis, genes of like effects must be completely associated with the parents. Therefore, selection of parents contrasting for the trait being measured is crucial for this type of investigation. Any dispersal of like genes among the two parents may cause cancelling because of some effects, resulting in the underestimation of additive (*d*), additive × additive (*i*) and additive × dominance (*j*) effects. Since the study was undertaken using parents contrasting mainly for grain iron and zinc concentrations, estimates of additivity for other traits could be underestimated. Dominance effects (*h*) are not influenced by the distribution of the alleles in the parents and estimates should be accurate (Mather and Jinks, 1982). From the breeder's point of view, having dominance as major type of gene action for the most important traits suggested that selection for these traits would be quite difficult and a long-term process.

Plant height and grain yield showed different results in the two crosses studied. Hence there is a need for further study using more contrasting parents to understand the inheritance of these traits. Dominance (*h*) and dominance × dominance (*l*) effects for all the traits studied took opposite signs suggesting the involvement of duplicate type of non-allelic interaction in controlling these traits. Plant height, in the cross IS13205 × IS23464, had positive dominance (*h*) and dominance × dominance (*l*) components which indicates the role of complementary type of epistasis. The component dominance (*h*) was positive and dominance × dominance (*l*) was negative for almost all the traits in the two crosses which revealed the involvement of duplicate interaction between increasing genes, whereas for plant height in the cross IS13205 × SPV1359 reverse was found i.e., component dominance (*h*) was negative and dominance × dominance (*l*) was positive which showed duplicate interaction between decreasing genes (Hayman and Mather, 1955). This duplicate epistasis caused a higher degree of reduction of the positive effects of dominant genes, leading to smaller yield. This kind of epistasis generally hinders the improvement through selection and hence, a higher magnitude of dominance and dominance × dominance type of interaction effects would not be expected. It is also indicated that selection would have to be deferred till later generations until a high level of gene fixation is attained. Subsequent intermatings between promising lines may be important in accumulating favorable genes. Complementary epistasis for plant height for the cross IS13205 × IS23464 facilitates the exploitation of heterosis for this character in this particular cross.

From the results obtained in the present investigation it can be concluded that there was no clear-cut relationship among estimated genetic effects for the traits. This relationship can be disturbed either by inappropriate (insufficient) sample size and/or structure or by higher order and complex gene interactions. Considering that these results are based on digenic interaction model using only two crosses, there is a need for further study using crosses developed from contrasting parents to fit a trigenic interaction and linkage model. Such crosses besides revealing precise genetic information on the inheritance of trait, contributes for a strong breeding programme to develop desirable genotypes.

Chapter V

SUMMARY AND CONCLUSIONS

Chapter V

SUMMARY AND CONCLUSIONS

Sorghum is an important food and feed crop in the semi-arid regions of the world where it is grown predominantly under rainfed conditions. Biofortification, enhancing the micronutrient concentrations by genetic means, is an effective approach to combat micronutrient malnutrition prevailing in the world today. Biofortifying sorghum can help to address the malnourished poorer sections of the world mainly consuming sorghum at a cheaper price. Hence, the present investigation entitled "Genetic analysis of grain iron and zinc concentrations in sorghum (*Sorghum bicolor* (L.) Moench)" was undertaken at the ICRISAT farm, Patancheru, Hyderabad using generation mean analysis with two crosses (IS13205 × SPV1359 and IS13205 × IS23464) generated using parents contrasting for grain iron and zinc concentrations with the aim of obtaining information on gene action governing grain iron and zinc concentrations, estimating the genetic parameters, determining the correlation of grain iron and zinc concentrations with grain yield and other important traits in sorghum. From each cross six generations *i.e.*, P_1, P_2, F_1, F_2, B_1 and B_2 were generated and were analyzed in an experimental trial conducted in post-rainy season, 2012-13 using Randomized Complete Block Design with three replications. Data were recorded for days to 50% flowering, plant height, panicle length and width, grain yield plant^{-1}, plant aspect score, 100-grain weight, and grain iron and zinc concentrations using Inductively Coupled Optical Emission Spectromety (ICP-ES).

Analysis of variance was done to test the significance of differences between the generations and genetic parameters like Phenotypic Coefficient of Variation, Genotypic Coefficient of Variation, heterosis, residual heterosis, inbreeding depression, heritability in broad sense and narrow sense, genetic advance as a percentage of mean and degree of dominance were estimated. Generation mean analysis was carried out using the scaling tests given by Mather (1949) and Hayman and Mather (1955) and six parameter model given by Hayman (1958). Correlation coefficients were calculated at phenotypic level using the formulae suggested by Falconer (1981).

The analysis of variance showed significant differences among generations for all the traits studied except for days to 50% flowering in the cross IS13205 × SPV1359, for 100-grain weight in the cross IS13205 × IS23464 and for panicle width in both the crosses for which generation mean analysis had not been done. PCV% ranged from 9.90

for panicle width in the cross IS13205 × SPV1359 to 49.28 for grain yield in the cross IS13205 × IS23464. GCV% ranged from 2.49 for days to 50% flowering to 36.92 for grain yield in the cross IS13205 × SPV1359. The phenotypic coefficient of variation was moderately higher than genotypic coefficient of variation for all the traits under study except days to 50% flowering, which suggested moderate influence of environment on these traits. PCV% and GCV% estimates were comparatively lesser for grain iron and zinc concentrations which indicated lesser variability in the material used. Moreover, there is much difference between PCV% and GCV% estimates of grain iron and zinc concentrations which highlights the importance of environment on the accumulation of these micronutrients. Heritability in broad sense varied from 4.21 for days to 50% flowering to 88.1 for panicle length in the cross IS13205 × SPV1359. Comparatively grain iron concentration recorded higher heritability estimates than grain zinc concentration. Genetic advance as percentage of mean (GAM) ranged from 1.05 for days to 50% flowering to 65.84 for grain yield in the cross IS13205 × SPV1359. High heritability coupled with high genetic advance was observed in case of plant height, panicle length, and grain yield and grain iron and zinc concentrations which indicated the preponderance of additive genetic effect which makes the selection successful. However, heritability in narrow sense ranged from 2.27 for grain iron concentration in the cross IS13205 × IS23464 to 19.62 for panicle length in the cross IS13205 × IS23464 which was low for all the traits studied which signifies the lesser contribution of additive gene action. More clarity on this can be obtained from generation mean analysis.

In the present material, average heterosis ranged from -4.73 for 100-grain weight in the cross IS13205 × SPV1359 to 25.37 for grain iron concentration in the cross IS13205 × SPV1359. Average heterosis for grain iron and zinc concentrations were positively significant in both of the crosses studied. Residual heterosis over mid-parent ranged from 3.91 for 100-grain weight for the cross IS13205 × SPV1359 to 38.18 for grain yield in the cross IS13205 × IS23464. Residual average heterosis for grain iron and zinc concentrations was significant in positive direction in the cross IS13205 × IS23464. Heterobeltiosis ranged from -1.85 for grain iron concentration in the cross IS13205 × SPV1359 to 12.15 for panicle length in the cross IS13205 × IS23464. Lack of heterobeltiosis indicated that breeding hybrids with high grain iron and zinc densities would require high levels of these micronutrients in both parental lines. Residual heterosis over better parent ranged from 2.78 for panicle length in the cross IS13205 × SPV1359 to -55.55 for grain yield in the cross IS13205 × IS23464. Inbreeding depression varied from -0.46 for days to 50% flowering in the cross IS13205 × IS23464 to 56.07 for grain yield in the cross IS13205 × SPV1359. Degree of dominance for all the traits was more than one which indicated the role of over dominance.

Correlation analysis revealed that panicle length was correlated significantly in positive direction with panicle width, grain yield plant^{-1} and grain zinc concentration. Grain iron concentration showed highly significant positive correlation with grain zinc concentration in the cross IS13205 × SPV1359. Grain yield plant^{-1} showed significant negative correlation with 100-grain weight while grain iron concentration showed highly significant positive correlation with grain zinc concentration in the cross IS13205 × IS23464. The results indicated that it is feasible to select simultaneously for grain iron and zinc concentrations in sorghum and there is no penalty on yield and other agronomic traits while improving these micronutrients.

Generation mean analysis revealed that atleast one (for grain yield plant^{-1}, 100-grain weight and grain iron concentration in the cross IS13205 × SPV1359 and for grain yield plant^{-1} and grain iron concentration in the cross IS13205 × IS23464) to maximum of three scaling tests (plant height and panicle length in the cross IS13205 × SPV1359) were significant for the traits studied except days to 50% flowering, where all the scaling tests were non-significant, which indicated the presence of non-allelic interactions. Hayman's six parameter model showed high significance for the estimated values of mean effects (*m*) indicating that all the studied traits in two crosses were quantitatively inherited. Additive component was significant for plant height in negative direction and for grain yield plant^{-1} and grain zinc concentration in positive direction in the cross IS13205 × SPV1359 and for plant height in positive direction and for grain yield plant^{-1} in negative direction in the cross IS13205 × IS23464. Dominance effects were significant for plant height and grain zinc concentration in negative direction and panicle length and grain yield plant^{-1} in the cross IS13205 × SPV1359 and for panicle length in the cross IS13205 × IS23464. Among interactions additive × additive was significant for all the traits except 100-grain weight in the cross IS13205 × SPV1359, while in the cross IS13205 × IS23464, it was significant for almost all the traits studied except plant height. Additive × dominance was significant for plant height, grain yield plant^{-1}, 100-grain weight and grain iron concentration in the cross IS13205 × SPV1359, while in the cross, IS13205 × IS23464, it was significant for panicle length, grain iron and zinc concentrations. These components were significant in negative direction, only for grain iron and zinc concentrations, whereas for other traits they were positively significant in both the crosses. Dominance × dominance was significant for all the traits studied, except 100-grain weight in the cross IS13205 × SPV1359, where its significant value is in negative direction for panicle length and grain yield plant^{-1}. This component was negatively significant only for panicle length in the cross IS13205 × IS23464. Higher magnitude of dominance component over additive component was observed for almost all the traits studied in both the crosses. Among interactions a minimum of one (for 100-grain weight in the cross IS13205 × SPV1359 and plant height and grain yield plant^{-1} in the cross IS13205 × IS23464) to maximum of three (plant height and grain yield plant $^{-1}$ in the cross IS13205 × SPV1359) were significant with higher magnitude of dominance × dominance followed by additive × dominance and additive × additive. The interaction effects are of higher value compared to direct effects. The signs of dominance (*h*) and dominance × dominance (*l*) were opposite in most of the cases indicating duplicate type of epistasis except for plant height in the cross IS13205 × IS23464. The sign of dominance (*h*) was negative and dominance × dominance (*l*) was positive in most of the cases (particularly grain iron and zinc concentrations) which indicated the existence of duplicate epistasis between decreasing genes.

Conclusion and future strategy:

The PCV% and GCV% differed by moderate values which depict the moderate role of environment in trait expression. But for grain iron and zinc concentrations, the gap is wider indicating the effect of environment in trait expression. High heritability coupled with high genetic advance as percentage of mean was observed for most of the traits studied which indicated the predominant role of additive gene action. Heterosis over mid parent and better parent was significant mostly in negative direction which revealed that the performance of hybrid was more towards the poorer parent for most of the traits which, of course, is desirable for plant height and days to 50% flowering. High magnitude of heterosis coupled with high inbreeding depression in some cases while in remaining cases their magnitudes were in opposite directions. Hence, there is a need to study these parameters using more number of crosses to arrive at valid conclusions. Highly significant positive correlation between grain iron and zinc concentrations strengthened the possibility of simultaneous improvement of grain iron and zinc concentrations. Lack of correlation of grain yield plant^{-1} and 100-grain weight with grain iron and zinc concentrations indicated that the breeding for grain iron and zinc concentration doesn't lead to reduction in yield and grain size. However, there is need to confirm these results using different types of populations.

In general, both additive and non-additive gene effects appear to be effective for all the traits studied. Therefore, improvement of these traits appears to beset with difficulties as simple selection techniques will not be able to fix superior lines in the early segregating generations. Postponement of selection of superior lines to later generations in pedigree breeding will be effective. However, one or two cycles of recurrent selection followed by pedigree breeding will be effective and useful to utilize both additive and non-additive gene effects. Predominance of dominance and epistatic interactions in both the crosses for iron and zinc indicates that selection for higher grain iron and zinc concentrations would be more effective if the dominance and epistatic effects are first reduced by a few generations of selfing. Moreover, a significant gap between PCV and GCV percentages demand a multi-location and multi-seasonal experimentation to test the consistency of results obtained.

Though generation mean analysis highlights the predominance of dominance and dominance × dominance interactions, significant estimates of positive heterosis is obtained in few cases which might be due to the duplicate gene action which defeats the heterotic effects of the dominant genes. The conclusions drawn from the gene effects for different traits are based on digenic interaction model and in two crosses only. However, possibilities of trigenic or higher order interaction and/or linkages among the interacting genes cannot be ruled out. Hence, there is a need for further study with more number of generations to fit a trigenic interaction and linkage model using crosses involving parents contrasting for the respective traits.

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