Original Research Article

Genetic Insights for Nutritional Traits in Elite Rice (*Oryza sativa* L.) Crosses Using Generation Mean Analysis

Abstract:

In the present experiment, the fitting of digenic interaction model indicates the prevalence of interaction effects especially additive x additive [i] type in addition to the main additive and dominance gene effects in case of amylose content. A duplicate type of epistasis was registered in the second cross (WGL-32100 x RP-Bio-5478-166) as the signs were in opposite sides. For protein content, two crosses (MTU 1010 x NH-686 and WGL-32100 x RP-Bio-5478-166), registered duplicate type interaction. The overall genetic analysis in the three crosses suggests a simple pedigree method to develop desirable lines from only cross *viz.*, RP-Bio-5478-185 x NH-686 would be feasible. The generation mean analysis clearly indicated that apart from the additive and dominance gene effects, significant magnitudes [i] type were observed to play greater role in expression of iron content in rice. In comparison to the estimates of [d], the component due to dominance genetic effects [h] is very high in all the three crosses uniformly. But, [i] type of epistasis which is fixable in nature was highly significant giving a way for direct selection. However, due to prevalence of [h] component significantly in all the crosses, inter mating in early segregating generations, would further increase chances to develop iron enriched homozygous lines. Studies for zinc content revealed that among the interactions, [i] and [j] types were found to be significant, whereas the [l] component was totally negligible. When fixable components are taken into consideration, the [i] type (additive x additive) was observed to be more predominant in desirable side in all the three crosses. From the study, all the three crosses were considered as superior on one or other way and among these three crosses, the last one (RP-Bio-5478-185 x NH-686) was considered as superior one.

Key words: Rice, Generation mean analysis, Digenic interaction model, $\chi 2$ test, Epistasis

Introduction:

Research efforts focused on development of high-yielding, disease & pest resistant and quality varieties and adoption of modern production technologies resulted in enhanced production leading to self-sufficiency in the country. Application of biometrical procedures in plant breeding in recent years led to a greater understanding of genetics of quantitative traits such as yield and proved to be useful to plant breeders for genetic analysis and planning for sound breeding programme. Along with yield, grain and nutritional quality has also become a primary consideration in rice breeding programs not only in India but also in various rice growing countries across the world. Grain quality characteristics are very important in rice breeding as it is predominantly consumed as a whole grain. Like yield, grain quality is not easily amenable to selection due to its complex nature. Lack of clear cut perception regarding the component traits of good quality rice is one of the important reasons for the tardy progress in breeding for rice varieties with fine grain quality. The quality characters of rice include physical attributes like grain appearance, kernel length/breadth ratio, milling parameters like hulling, milling percentages, head rice recovery etc. cooking quality and eating qualities like elongation ratio, gelatinization temperature. Now, nutritional elements like protein, iron and zinc contents were included as important quality attributes in view of their role in human health maintenance. Genetic variation for micronutrients in rice was studied and reported for iron and zinc (Gregorio et al., 2000; Zhang et al., 2004). Iron and zinc contents in brown and milled rice of national and international germplasm need to be estimated for the identification of donors for future deployment in the

nutritional breeding programs. Incorporation of these characters into high yielding semi dwarf varieties are essential in this era of quality breeding.

The ability of parents to combine well depends upon complex interaction among genes, which cannot be predicted from yield performance and adaptability of the parents (Allard, 1960). Most of the reports for gene action in rice are based on the diallel mating which does not provide information regarding non-allelic gene actions. The non-allelic gene actions could inflate the measures of additive and dominance components. Estimation of gene effects based on generation mean analysis provides requisite information to formulate the breeding strategy. Thus, generation mean analysis is a useful technique in plant breeding for estimating main gene effects (additive and dominance) and their digenic (additive x additive, additive x dominance, and dominance x dominance) interactions responsible for inheritance of quantitative traits. It helps us in understanding the performance of the parents used in crosses and potential of crosses to be used either for heterosis exploitation or pedigree selection. The nature of inheritance and type of gene action governing quality traits are many and complex and when genetically manipulated to some extent paved the way for the success in quality improvement through conventional breeding methods. Hence a better understanding of the factors that contribute to the overall grain quality of rice will lay the foundation for developing new breeding and selection strategies for combining high quality with high yield. Genetic enhancement of key food crops with enhanced nutrients is advocated as the most promising approach to address the problem of malnutrition (Graham et al., 2001; Bouis, 2002) which can be possible with understanding the genetic analysis among quality and nutrional traits in rice.

Considering the fact that nutrient traits are the most important complex traits and that their improvement is the most frequent goal of rice breeding programs. Keeping this aspect in view, the present investigation was undertaken to study the gene action for nutrient traits in rice.

Materials and Methods

The present investigation was carried out at the Regional Agricultural Research Station, Warangal, Telangana State, India which is located at an altitude of 304 M above MSL, 17.97° N latitude and 79.60° E longitude during two years 2014-15 and 2015-16 utilizing both *Kharif* and *Rabi* crop seasons in each year. The main crop seasons in Telangana State, India can be called as rainy (June-Dec) and post rainy (Nov-April) seasons.

Development of material for Generation Mean Analysis

Five parents *viz.* MTU 1010, WGL-32100, RP-Bio-5478-185, NH-686 and RP-Bio-5478-166 were selected based on contrasting characters and developed material (F_1, F_2, BC_1 and BC_2) for 3 independent crosses (MTU 1010 X NH – 686, WGL-32100 X RP-Bio-5478-166 and RP-Bio-5478 -185 X NH-686) to study the presence of non allelic interactions through generation mean analysis for quality and nutritional characteristics. Variation for kernel dimensions (long slender x short bold, medium slender x short bold and short bold x short bold) and flowering duration was considered as criteria for selection of parents for generation mean analysis.

Generation of F_1 hybrids

Hybridization & crop management techniques

During *Kharif* 2014, five parents were transplanted each in four rows in a crossing block at spacing of 20 x 15 cm and 4 sets were maintained. Crosses were effected to produce three F_1 s. The selected parents for study of generation mean analysis were sown at staggered intervals of seven days to facilitate continuous availability of pollen during crossing. Twenty eight days old seedlings were transplanted at spacing of 30 x 15 cm. Hybridization was done by clipping method of emasculation as suggested by Jennings *et al*. (1981). Spikelets were emasculated in the afternoon and pollination was done in morning of next day between 11 – 1.00 P.M. Adequate care was taken to produce sufficient seed required for studying different generations.

.**Study of Generations**

Three crosses (MTU 1010 X NH-686, WGL-32100 X RP-Bio-5478-166 and RP-Bio-7458-185 X NH-686) were selected wherein the parents had contrasting features for different traits under study. The F_1's generated from five parents during *Kharif*, 2014, and corresponding F_2's (seed obtained from F_1 generation raised during successive *Rabi*, 2014-15 and crosses were selfed and backcrossed with their respective parents to obtain backcrosses (BC_1 and BC_2) generations respectively. Parents were also selfed to ensure 100% genetic purity. Thus, seed of six basic generations, P_1, P_2, F_1, F_2, BC_1 and BC_2 was in hand for these three crosses at the end of the season *Rabi* 2014-15.

The experimental material (P_1, P_2, F_1, F_2, BC_1, BC_2) was planted separately side by side for this study and data generated. The number of rows for P_1, P_2, F_1, F_2, BC_1 and BC_2 were 1, 1, 1, 8, 5, and 5 respectively. The mean data was used for final statistical analysis.

Estimation of Quality and Nutritional Traits

Amylose content (%)

Amylose (%) content in the rice sample was estimated as per the procedure given by Jennings *et al*. (1979).

Crude protein content (%)

Protein content (%) = N content x 5.65

Nitrogen content of the rice grain was estimated by following micro-kjeldahl method (A.O.A.C, 1980).

Grain iron concentration zinc concentration (ppm)

Grain iron and zinc concentrations were determined by X-Ray fluorescence Spectrometry (XRF) (EDXRF, model-X-supreme8000).

Statistical Analysis

Generation means and gene effects

The concept of Generation Mean Analysis (GMA) was developed by Hayman and Mather (1955) and Jinks and Jones (1958) for the estimation of genetic components of variation (m, d, h, I, j, l). This technique involves six different generations *viz.*, parents (P_1 and P_2), their F_1, F_2 and back crosses (BC_1 and BC_2). Accordingly, the means were computed for each generation of P_1, P_2, F_1, F_2, BC_1 and BC_2 for each cross over three replications in the present study. 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.

Joint Scaling test

Data were subjected to joint scaling test (Cavalli, 1952) in view of the advantages associated with this test. The Cavalli's joint scale test has two distinct advantages over scaling tests of Mather, 1949. Firstly, this test combines very effectively several scaling tests into one utilizing the entire available 6 generation mean and offers a more genetical approach. Secondly, this test provides best possible estimates of all the parameters needed to describe differences among the means of families, in addition to the test of adequacy of additive-dominance model. In the present study, the characters which fail to show significant variation among generations in any cross was not subjected to further genetic analysis of generation means. As only 6 generations are available in the event of significance of 6 parameters (m, d, h, i, j, l) and significance of χ^2 test also further analysis cannot be carried out and the result of χ^2 test cannot be interpreted due to non availability of degrees of freedom. Fortunately, in the present study, there were certain components which were non-significant, these were deleted and further analysis was carried out and χ^2 test was done using left over degrees of freedom.

Initially we tried a model with just 'm' first , if this adequately explained the variations in the trait , there was no need to proceed further to estimate any other genetical parameters, otherwise next higher parameters like d, h *etc.*, were introduced until χ^2 test value become non significant.(Kearsey and Ponni, 1996).

First it was fitted into 3 parameter model, when $\chi 2$ value was significant, it indicated that data does not fit into additive - dominance model, hence, sequential model was followed. The data was first subjected to 6 parameter model, and when all the components were significant in six

parameter model, no further analysis was carried out. In the event of non significance of any one o the components, the same was deleted and data fitted into 5 parameter model. This procedure was followed until the χ^2 value is non significant and all the components were significant in the model. Fitness in 5 or 4 parameter model indicated the presence of digenic non allelic interaction.

The following procedure was followed to estimate the components in the joint scaling test. The parameters m, [d] and [h] estimated from the observed mean of the available types of generations were compared with expected values derived from the estimates of these three parameters. The six equations which are obtained by equating the observed family means to their expectations in terms of m, [d], [h], [i], [j] and [l] were used for estimating these parameters. Since, the number of equations is higher than the number of parameters to be estimated, least square technique was followed.

Results and Discussion

The mean and variances of mean values of different generations utilized for generation mean analysis with respect to grain quality and nutritional traits of three crosses are presented in tabular form (Table 1, 2 &3).

Generation mean analysis was carried out using 6 generations means (P_1, P_2, F_1, F_2, B_1 & B_2). The adequacy of additive – dominance model was tested by using A, B, C, D scales of Mather (1949) (Table 4,5 & 6).and also further confirmed by the Joint scaling test of Cavalli (1952) (Table 7). As the Joint scaling test is a comprehensive test of simple additive – dominance model replacing A, B, C, D scaling test and involves weighted regression analysis, the components would be estimated and tested with more precision. Hence, a sequential method was followed to identify the best fit model in which the all the possible components would be significant and at the same time the χ^2 test value becomes non significant. Major advantage of this test is that we can delete a non significant component in the sequential process and estimate the remaining significant components with maximum likeyhood precisions.

Initially we tried a model with just 'm' first , if this adequately explained the variations in the trait , there was no need to proceed further to estimate any other genetical parameters, otherwise next higher parameters like d, h *etc.*, were introduced until χ^2 test value become non significant.(Kearsey and Ponni, 1996).

In the present study, fortunately at least one component out of six was non significant, having at least one degrees of freedom to facilitate the χ^2 test. This procedure was adopted for three crosses uniformly and estimated the possible parameters by weighted least square method and discussed thoroughly (Table 7).

For amylase content, the fitting of digenic interaction model indicates the prevalence of interaction effects especially additive x additive [i] type in addition to the main additive and dominance gene effects. These findings are in agreement with the earlier reports of Mahalingam and Natarajan (2010) and Subbalaxmi *et al.* (2016). In the present study, both additive and dominance genetic effects were found to be significant. In addition, the [i] type of epistasis was also found to play a greater role in expression of amylase content in desirable direction. A duplicate type of epistasis was registered in the second cross (WGL-32100 x RP-Bio-5478-166) as the signs were in opposite sides. Keeping in view the highly significant estimates of fixable, additive x additive type of genetic variation coupled with existence of higher magnitudes of additive gene effects, a better scope is expected for isolation of promising genotypes through simple selection procedures. However, attempting few crosses among the selected segregating genotypes and postponing the selections for some time may further improve the breeding efficiency (Ramli *et al.*, 2016).

For protein content, significant values of 'm' were estimated in the crosses studied. In two crosses (MTU 1010 x NH-686 and WGL-32100 x RP-Bio-5478-166) the interaction effect was observed which was of duplicate type as the [h] and [l] components showed opposite signs. The genetic variation was largely controlled by dominance gene effects of negative type in these two crosses, thus a little scope was indicated for improvement of protein content for these two crosses. In view of the dominance gene effects on negative side and lowering of desirable interaction

effects due to mutual cancellation, heterosis breeding would not be profitable. The overall genetic analysis in the three crosses suggests a simple pedigree method to develop desirable lines from only cross *viz.,* RP-Bio-5478-185 x NH-686.

Iron deficiency is probably the most wide spread micro nutrient deficiency in rural areas, where rice is the staple food. Therefore, apart from breeding, the approaches like genetic engineering, biochemical and physical have been frequently used as prospective methods to regulate iron content and bio availability in rice grains. This present study was envisaged to understand the genetics of iron content as little efforts were made so far in these lines. The generation mean analysis clearly indicated that apart from the additive and dominance gene effects, significant magnitudes of digenic interactions especially [i] type were observed to play greater role in expression of iron content in rice. In one cross, RP-Bio-5478-185 x NH-686, all the types of digenic interactions were present with duplicate dominant epistasis. In comparison to the estimates of [d], the component due to dominance genetic effects [h] is very high in all the three crosses uniformly. It is interesting to note that [i] type of epistasis which is fixable in nature is highly significant giving a way for direct selection. However, due to prevalence of [h] component significantly in all the crosses, inter mating in early segregating generations, would be further useful to develop iron enriched homozygous lines.

Apart from iron, zinc is another important element to be consumed at optimum level as per the dietary requirements. Therefore, in most of the programmes both iron and zinc would be considered for simultaneous improvement. To understand the genetic architecture of zinc content in rice, the generations mean analysis was carried out and fitted in various models. The first cross combination was fitted in digenic interaction 4 parameter model, whereas, other two crosses were fitted at 5 parameter level. Among the interactions, [i] and [j] types were found to be significant, whereas the [l] component was totally negligible. When fixable components are taken into consideration, the [i] type (additive x additive) was observed to be more predominant in desirable side in all the three crosses, whereas [d] type was significant only in one cross *i.e* RP-Bio-5478-185 x NH-686, hence all the three crosses were considered as superior on one or other way. This findings is in agreement with the earlier report of Maddeppa Mallimar *et al.,(*2017). The dominance effects were prevalent in desirable direction in all the crosses where as the additive component was significantly negative in first and second crosses. In view of the predominant role of dominance effects and also the fixable interaction effects, the breeding methods like recurrent selection, biparental mating would be highly advantageous and among these three crosses, the last one (RP-Bio-5478-185 x NH-686) was considered as superior one.

Statistical analyses

Means of three experimental values were used. Entire analyses were carried out by using Indostat available at Professor Jayashankar Telangana State Agricultural University, Hyderabad.

Table 1. Estimates of generation mean and variances of mean for nutrient characters for the cross MTU 1010 x NH-686

Character	MTU 1010 x NH-686											
	P_1		P_2		F_1		F_2		BC_1		BC_2	
	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean
Amylose content (%)	23.02 ±0.23	0.05	20.94 ±0.33	0.10	24.89 ±0.22	0.05	20.76 ±0.18	0.03	22.68 ±0.44	0.19	22.06 ±0.86	0.74
Protein content (%)	7.40 ±0.22	0.04	7.54 ±0.16	0.02	9.64 ±0.21	0.04	7.66 ±0.47	0.22	7.51 ±0.17	0.03	7.76 ±0.23	0.05
Iron content (ppm)	9.80 ±0.02	0.007	10.03 ±0.12	0.01	10.02 ±0.24	0.05	9.17 ±0.07	0.005	9.46 ±0.07	0.006	9.79 ±0.08	0.007
Zinc content (ppm)	11.11 ±0.06	0.004	13.76 ±0.18	0.03	13.00 ±0.16	0.02	10.78 ±0.69	0.48	11.26 ±0.42	0.17	13.00 ±0.29	0.08

Table 2. Estimates of generation mean and variances of mean for nutrient characters for the cross WGL-32100 X RP-Bio-5478-166

Character	WGL-32100 X RP-Bio-5478-166											
	P_1		P_2		F_1		F_2		BC_1		BC_2	
	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean
Amylose content (%)	22.68 ±0.27	0.07	19.68 ±0.66	0.43	22.77 ±0.16	0.02	21.19 ±1.41	1.98	21.51 ±0.36	0.13	19.53 ±0.61	0.37
Protein content (%)	7.11 ±0.06	0.004	8.53 ±0.26	0.07	8.88 ±0.06	0.004	7.98 ±0.54	0.29	10.82 ±0.37	0.01	8.12 ±0.27	0.07
Iron content (ppm	10.75 ±0.41	0.17	12.89 ±0.20	0.04	11.99 ±0.21	0.04	9.91 ±0.17	0.03	14.30 ±0.39	0.14	10.78 ±0.37	0.14
Zinc content (ppm)	14.44 ±0.11	0.01	19.69 ±0.16	0.02	22.69 ±0.22	0.05	14.15 ±0.28	0.08	14.30 ±0.39	0.15	20.81 ±0.47	0.22

Table 3. Estimates of generation mean and variances of mean for nutrient characters for the cross RP-Bio-5478 -185 x NH-686

Character	RP-Bio-5478 -185 x NH-686											
	P_1		P_2		F_1		F_2		BC_1		BC_2	
	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean	Mean	Variance of mean
Amylose content (%)	19.36 ±0.40	0.16	21.36 ±0.25	0.06	21.01 ±0.45	0.20	19.29 ±0.30	0.09	19.66 ±0.26	0.07	21.13 ±0.44	0.19
Protein content (%)	7.84 ±0.16	0.02	7.56 ±0.15	0.02	8.36 ±0.20	0.04	7.80 ±0.42	0.18	7.97 ±0.07	0.005	7.79 ±0.13	0.01
Iron content (ppm)	11.64 ±0.23	0.05	9.85 ±0.20	0.04	15.32 ±0.35	0.12	10.15 ±0.52	0.27	10.70 ±0.28	0.07	14.01 ±0.24	0.06
Zinc content (ppm)	21.31 ±0.34	0.12	13.78 ±0.22	0.05	19.15 ±0.28	0.08	14.44 ±0.25	0.06	14.44 ±0.26	0.07	18.34 ±0.37	0.14

Table 4. Scaling tests of generation means for the cross MTU-1010 x NH-686 for nutrient traits

S. No	MTU-1010 x NH-686				
	Character	Scales			
S. No		A	B	C	D
1	Amylose content (%)	-2.55* ± 0.94	-1.70 NS ± 1.76	-10.72** ± 0.94	-3.23** ± 1.03
2	Protein content (%)	-2.01** ± 0.46	-1.65 **± 0.54	-3.57 NS ± 1.98	0.04 NS ± 1.00
3	Iron content (ppm)	-0.90** ± 0.29	-0.47NS ± 0.32	-3.17** ± 0.57	-0.89** ± 0.18
4	Zinc content (ppm)	-1.51 NS ± 0.86	-0.75 NS ± 0.63	-7.75** ± 2.81	-2.74 NS ± 1.48

*Significant at 5 % level, ** Significant at 1 % level

Table 5. Scaling tests of generation means for the cross WGL-32100 X RP-Bio-5478-166 for nutrient traits

S. No	WGL-32100 X RP-Bio-5478-166				
	Character	Scales			
		A	B	C	D
1	Amylose content (%)	-2.42** ± 0.79	-3.38** ± 1.40	-3.14 NS ± 5.69	1.33 NS ± 2.90
2	Protein content (%)	-1.80** ± 0.27	-1.17 NS ± 0.62	-1.50 NS ± 2.19	0.73 NS ± 1.12
3	Iron content (ppm)	-1.10 NS ± 0.88	-3.32** ± 0.81	-7.97** ± 0.93	-1.77* ± 0.63
4	Zinc content (ppm)	-8.53** ± 0.82	-0.75 NS ± 0.98	-22.89** ± 1.25	-6.80** ± 0.84

*Significant at 5 % level, ** Significant at 1 % level

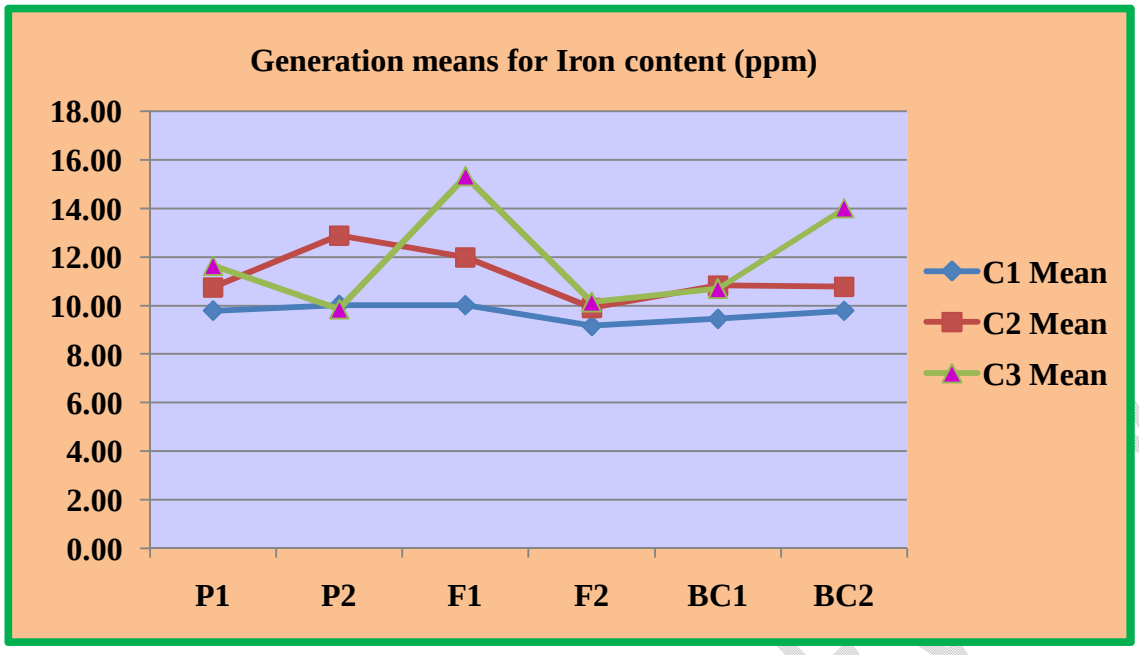
Table 6. Scaling tests of generation means for the cross RP-Bio-5478 -185 x NH-686 for nutrient traits

S. No	RP-Bio-5478 -185 x NH-686				
	Character	Scales			
		A	B	C	D
1	Amylose content (%)	-1.04NS ± 0.81	-0.10NS ± 1.02	-5.58** ± 1.60	-2.22** ± 0.80
2	Protein content (%)	-0.25NS ± 0.30	-0.34NS ± 0.37	-0.91NS ± 1.76	-0.15NS ± 0.86
3	Iron content (ppm)	-5.56** ± 0.70	2.85** ± 0.63	-11.52** ± 2.24	-4.40** ± 1.11
4	Zinc content (ppm)	-11.57** ± 0.70	3.75** ± 0.83	-15.61**± 1.22	-3.89** ± 0.68

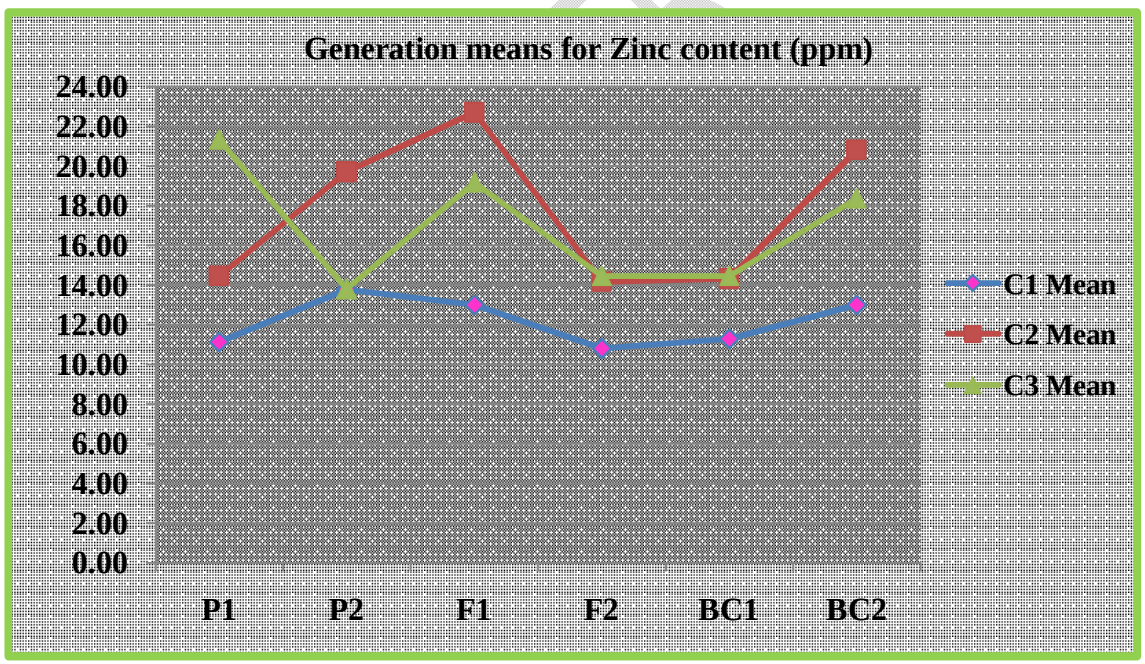
*Significant at 5 % level, ** Significant at 1 % level

Table 7. Genetic components of nutrient traits estimated through Joint scale test

Cross	Genetic components	Character			
		Amylose content (%)	**Protein content (%)**	**Iron content (ppm)**	**Zinc content (ppm)**
MTU 1010 x NH-686	m	16.64** ± 0.42	7.49** ± 0.13	8.29** ± 0.26	9.77** ± 0.84
	[d]	1.03** ± 0.19	-	-0.17** ± 0.05	-1.35** ± 0.09
	[h]	8.26** ± 0.57	-1.68* ± 0.69	1.79** ± 0.43	3.24**± 0.91
	[i]	5.35** ± 0.47	-	1.67** ± 0.26	2.68** ± 0.86
	[j]	-	-	-	-
	[l]	-	3.84** ± 0.73	-	-
χ^2 value/probability		**0.327(0.849) NS**	**1.026/(0.795) NS**	**2.763/(0.251) NS**	**1.711/(0.425) NS**
Model fitted		**4 parameter**	**3 parameter**	**4 parameter**	**4 parameter**
Type of epistasis		**-**	**duplicate**	**-**	**-**
WGL-32100 x RP-Bio-5478-166	m	21.12** ± 0.34	7.87** ± 0.12	7.80** ± 0.40	5.94** ± 0.58
	[d]	1.59** ± 0.32	-0.76**± 0.12	-0.90** ± 0.21	-2.62** ± 0.09
	[h]	-3.54* ± 1.49	-2.31** ± 0.53	4.17** ± 0.54	16.80** ± 0.71
	[i]	-	-	4.10** ± 0.46	11.13** ± 0.59
	[j]	-	-	-	-7.45 **±1.23
	[l]	5.19** ± 1.36	3.32** ± 0.50	-	-
χ^2 value/probability		**0.519/ (0.771) NS**	**1.542/ (0.463) NS**	**3.849/ (0.146) NS**	**2.446/ (0.116) NS**
Model fitted		**4 parameter**	**4 parameter**	**4 parameter**	**5 parameter**
Type of epistasis		**duplicate**	**duplicate**	**-**	**-**
RP-Bio-5478 -185 x NH-686	m	17.17** ± 0.75	7.66** ±0.10	-	9.73** ± 0.55
	[d]	-1.02** ± 0.21	0.57** ± 0.20	0.89** ± 0.15	3.76 ** ± 0.20
	[h]	3.48** ± 1.09	-	23.58** ± 0.80	9.41** ± 0.75
	[i]	2.67** ± 0.79	-	10.73** ± 0.15	7.81** ± 0.36
	[j]	-	-	-8.38* ± 0.80	-15.33** ± 0.98
	[l]	-	-	-8.26** ± 1.00	-
χ^2 value/probability		**1.815/(0.404) NS**	**3.548/ (0.471) NS**	**0.744/ (0.388) NS**	**0.000/ (0.989) NS**
Model fitted		**4 parameter**	**2 parameter**	**5 parameter**	**5 parameter**
Type of epistasis		**-**	**-**	**duplicate**	**duplicate**



C1: MTU 1010 x NH-686, **C2**: WGL – 3200 x RP-Bio-5478-166, **C3**: RP-Bio-5478 -185 x NH – 686
Figure No.1. Mean values of 6 Generations of 3 crosses for **Iron content**



C1: MTU 1010 x NH-686, **C2**: WGL – 3200 x RP-Bio-5478-166, **C3**: RP-Bio-5478 -185 x NH – 686
Figure No.2. Mean values of 6 Generations of 3 crosses for **Zinc content**

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