

Research Article GENERATION MEAN ANALYSIS FOR SHEATH BLIGHT DISEASE RESISTANCE AND YIELD-RELATEDTRAITS IN RICE (*Oryza sativa L*)

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Abstract: The In order to study the mode of gene action for sheath blight resistance and yield related traits a cross was made. Five populations viz., P₁, P₂, F₁, F₂, and F₃ were derived from the cross between high yielding susceptible rice variety 'Swarna sub-1' and resistant line 'Tetep'. The sheath blight susceptible high yielding variety Swarna sub-1 showed high disease severity (60.46%) compared with resistant parent Tetep (17.72%) whereas intermediate disease severity was observed in F₁ and three segregating populations. Among F₁, F₂ and F₃ population, F₁ showed less disease severity (20.09%) than F₂ and F₃ populations. The Swarna sub-1 recorded higher grain yield per plant compared with Tetep while the F₁ yielded more grain yield compared with the donor parent but less than the recurrent parent, but in the two segregating populations (F₂ and F₃), grain yield per plant were intermediate than non-segregating generations. All the traits related to yield as well as sheath blight resistance were significant in either one of the scales or in combination representing the existence of epistatic interactions between the genes involved. The dominance (h) and dominance × dominance (l) gene effects displayed opposite sign for the traits number of reproductive tillers per plant, plant height, days to maturity, length and breadth ratio after cooking and gel consistency indicating duplicate epistasis while complementary for days to heading, panicle length, weight of panicle, number of spikelets per panicle, test weight, yield per plant, length and breadth ratio before cooking, amylose content and per cent disease severity.

Keywords: Sheath blight resistance, Percent of disease severity, Segregation populations, Gene effects, Epistasis, Rice, Grain yield, Amylase content

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Introduction

Rice is a basic food for millions of people, having considerable importance in food and nutritional security. It is the second most widely consumed food grain in the world next to wheat. In India, Rice is a major crop and is the essential food for people in the most parts of the country. India has second place in rice production after China, accounting for 21% of world rice production. Sheath blight disease caused by pathogen Rhizoctoniasolani Kühn is one of the most significant diseases of rice, causing in massive yield loss in rice every year. It has been reported to cause 20-30% yield loss depending on the severity of infection and approximately 50% yield reduction in test plots of susceptible rice cultivars [23]. Use of resistant cultivars is the most economical and environmentally sound strategy in managing sheath blight. The accurate measurement of sheath blight resistance under field conditions depends on a range of environmental factors [4] and plant morphological traits, such as plant height [32,20], which interact, resulting in the observed variation in resistant/susceptible phenotypes. None of the rice lines resistant to sheath blight has been recognized till now. However, in some rice lines a significant degree of resistance to R. solani has been reported. An Indica rice line, Tetep, is a well recorded source of durable and broad spectrum resistance to rice blast in addition to quantitative resistance to sheath blight [1].

Genetic nature of sheath blight has been found to be complex and contentious issue in the earlier studies. There were some reports about the major gene conferring resistance to sheath blight. Non allelic dominant major resistance gene was also documented in resistant cultivars *viz*. Jasmine and Teqing [16]. On the opposite, genetics studies on the quantitative resistance to *R. solani* in rice have exhibit both multiple gene and major gene inheritance [24,10,32].

Because of the complication of the resistance response and lack of detailed knowledge about the loci associated, breeding efforts to increase the resistance to R. solani have been mostly unsuccessful [10]. The expression of trait is affected not only by large number of genes governing them but also by environmental effect. Frequently, these genes interact with each other causing distortions in Mendelian ratios and leading to novel phenotypes [19]. The estimation of epistasis assumes more significance in view of these 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. Generation mean analysis is an important statistical tool for identification of epistasis using various basic generations from a cross between two parents. To obtain the desired genetic improvement towards the development of better lines, it is crucial to collect information about genetic architecture of quantitative traits including grain yield. Therefore, the present investigation was undertaken to estimate the types of gene action of sheath blight resistance in rice, yield and yield contributing traits through generation mean analysis.

MATERIALS AND METHODS:

Plant Material and Experimental Design

The present work was carried out during *Kharif* 2011, 2012 and 2013 at Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi and at NRRI, Cuttack, Odisha. Experimental materials comprise of two rice genotypes including one submergence tolerance (susceptible to sheath blight disease) varieties (Swarna sub-1) and one sheath blight resistant genotype (Tetep).

Table-1 Mean performan	ce of five ae	neration m	aterials of the	cross Swarna	Sub-1×	Tetep for	fourteen traits
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P1±SEm	$P_2 \pm SEm$	F1±SEm	F ₂ ±SEm	F₃±SEm
12.07±0.57	8.80±0.59	9.93±0.49	9.23±0.31	9.33±0.34
96.33±1.26	129.07±1.31	135.20±1.61	137.61±1.87	134.80±1.53
140.93±0.94	103.53±0.50	125.87±1.31	122.25±1.42	120.33±1.34
103.33±0.46	72.27±0.43	91.27±0.98	89.75±0.84	90.00±0.86
25.84±0.68	26.42±0.73	28.73±0.53	27.05±0.47	26.55±0.71
4.08±0.09	3.85±0.18	4.9±0.10	4.07±0.06	4.15±0.08
223.13±6.95	191.20±5.32	214.07±8.75	208.23±	187.60±5.56
18.88±0.15	21.89±0.15	20.13±0.13	19.81±0.13	19.80±0.14
50.92±2.74	37.52±1.88	43.64±2.29	36.65±1.58	36.79±1.74
2.67±0.02	2.84±0.01	2.94±0.01	2.75±0.01	2.78±0.01
2.88±0.02	3.01±0.01	2.68±0.01	2.98±0.02	3.09±0.02
27.70±0.03	31.59±3.46	34.66±0.04	32.25 ±0.48	31.15±0.47
129.66± 0.42	120.77 ±034	124.92±0.31	121.50±3.25	126.03±3.56
60.46±1.32	17.72±0.81	20.09±1.15	37.00±1.46	38.49±1.48
	$P_{1\pm}SEm$ 12.07±0.57 96.33±1.26 140.93±0.94 103.33±0.46 25.84±0.68 4.08±0.09 223.13±6.95 18.88±0.15 50.92±2.74 2.67±0.02 2.88±0.02 27.70±0.03 129.66±0.42 60.46±1.32	$P_{1\pm}SEm$ $P_{2\pm}SEm$ 12.07 ± 0.57 8.80 ± 0.59 96.33 ± 1.26 129.07 ± 1.31 140.93 ± 0.94 103.53 ± 0.50 103.33 ± 0.46 72.27 ± 0.43 25.84 ± 0.68 26.42 ± 0.73 4.08 ± 0.09 3.85 ± 0.18 223.13 ± 6.95 191.20 ± 5.32 18.88 ± 0.15 21.89 ± 0.15 50.92 ± 2.74 37.52 ± 1.88 2.67 ± 0.02 2.84 ± 0.01 2.88 ± 0.02 3.01 ± 0.01 27.70 ± 0.03 31.59 ± 3.46 129.66 ± 0.42 120.77 ± 0.81	P1±SEmP2±SEmF1±SEm12.07±0.57 8.80 ± 0.59 9.93 ± 0.49 96.33±1.26129.07±1.31135.20±1.61140.93±0.94103.53±0.50125.87±1.31103.33±0.4672.27±0.4391.27±0.9825.84±0.6826.42±0.7328.73±0.534.08±0.093.85±0.184.9±0.10223.13±6.95191.20±5.32214.07±8.7518.88±0.1521.89±0.1520.13±0.1350.92±2.7437.52±1.8843.64±2.292.67±0.022.84±0.012.94±0.012.88±0.023.01±0.012.68±0.0127.70±0.0331.59±3.4634.66±0.04129.66±0.42120.77±0.8120.09±1.15	P1±SEmP2±SEmF1±SEmF2±SEm12.07±0.57 8.80 ± 0.59 9.93 ± 0.49 9.23 ± 0.31 96.33±1.26129.07±1.31135.20±1.61137.61±1.87140.93±0.94103.53±0.50125.87±1.31122.25±1.42103.33±0.4672.27±0.4391.27±0.9889.75±0.8425.84±0.6826.42±0.7328.73±0.5327.05±0.474.08±0.093.85±0.184.9±0.104.07±0.06223.13±6.95191.20±5.32214.07±8.75208.23±18.88±0.1521.89±0.1520.13±0.1319.81±0.1350.92±2.7437.52±1.8843.64±2.2936.65±1.582.67±0.022.84±0.012.94±0.012.75±0.012.88±0.023.01±0.012.68±0.012.98±0.0227.70±0.0331.59±3.4634.66±0.0432.25±0.48129.66±0.42120.77±0.8120.09±1.1537.00±1.46

Table-2 Scaling test, estimate of gene effects from analysis of generation mean for fourteen traits

Traits	Simple S	Scaling Test	Components of Generation Mean (5 parameter model)				Epistasis	
	С	D	m	d	h	i		
NTP	0.60±1.04	-18.67**±1.41	9.23**±0.18	0.63±0.24	-0.20±0.94	-0.10±0.88	0.80*±2.36	D
PH	-54.64**±4.83	-231.02**±5.17	137.61**±1.08	-16.37**±0.53	5.89**±3.79	-16.61**±3.60	-21.41**±10.88	D
DM	7.20**±3.67	-248.30**±3.98	122.25**±0.82	18.70**±0.31	7.52*±2.94	3.89±2.75	-0.58±8.36	D
DF	-0.87±2.28	-175.10**±3.27	89.75**±0.49	15.53**±0.18	0.34±2.31	-3.12±1.96	5.38±5.87	С
PL	1.55**±1.37	-53.24**±1.76	27.05**±0.27	-0.29±0.29	2.44*±1.19	-0.17±1.13	1.89±3.12	С
WP	1.67**±0.22	-7.67**±0.32	4.02**±0.04	0.12±0.06	0.24**±0.21	-0.69**±0.19	3.06**±0.51	С
SPP	89.53**±18.56	-455.6**±19.78	208.23**±3.21	15.97**±2.53	85.58**±14.23	38.68**±13.34	12.18**±38.86	С
TW	1.79**±0.36	-40.79**±0.65	19.81**±0.08	-1.51**±0.06	0.23*±0.44	0.49**±0.37	0.80**±1.05	С
YPP	29.13**±4.91	-88.17**±7.18	36.65**±0.92	6.70**±0.96	4.29**±4.89	4.88**±4.39	19.38**±12.06	С
L/B Ratio BC	0.40*±0.03	-5.44**±0.04	2.75**±0.01	-0.09*±0.01	0.05±0.03	-0.14±0.03	0.68*±0.07	С
L/B Ratio AC	-0.69*± 0.06	-5.67**± 0.07	2.98**±0.01	-0.06*±0.01	-0.49**±0.05	-0.23 ±0.05	0.24 ±0.14	D
AC	-0.41±2.28	-61.49**±2.07	32.25 ± 0.28	-1.94 ±1.00	4.54**±0.55	-0.48 ±1.21	0.54 ±2.21	С
GC	14.27*±7.51	-241.36**±3.91	121.50**±1.87	4.45 ±0.16	-9.80*±3.82	-9.51*±4.63	33.29**±15.06	D
% DS	-29.63**±3.73	-75.20**±4.31	37.00**±0.84	21.37**±0.45	-15.23*±3.11	3.76 ± 2.93	-37.1**6 ±8.66	Ċ

** and *: Significant at 1 and 5 percent level, respectively

Five generations (P₁, P₂, F₁, F₂ and F₃) from the cross (Swarna sub-1 × Tetep) were evaluated in this study. The F₁s were made at Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi (UP) during *Kharif*, 2011. The subsequent generation - F₂ required for the study was developed in off season 2012 at NRRI, Cuttack, Odisha. F₃ was again planted in the main season, *Kharif*, 2013 at Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi.



Fig-1 Range of percent disease severity in different generations of cross Swarna sub-1 × Tetep

The experiment was laid in a randomized complete block design with three replications during *Kharif* 2011, 2012 and 2013 crop season. The parental lines and F₁s; F₂s and F₃s were planted in 1, 2 and 3 rows each of 3 m long at spacing of 30×15 cm, respectively. Data were recorded on 10 plants in case of parents and F₁s, 30 plants of F₂s and 75 plants in F₃s per replication.

Pathogenicity test and Observations Recorded

Disease screening was done by artificial disease inoculation of Rhyzoctonia solani

(R.nagar strain) in the third leaf sheath from the top of the plant. Appearance of symptom was recorded 2 days after inoculation (DAI) and symptom lesion length and width were taken at 4^{th} , 8^{th} , 12^{th} and 16^{th} DAI intervals.



Plate 1- Reaction to sheath blight disease of the parents Swarna sub-1 and Tetep

The phenotypic traits were assessed on randomly selected plants from each individual entry in the segregating generations for nine quantitative traits *viz.*, number of productive tillers per plant (NTP), plant height (PH), days to 50 percent flowering (DF), days to maturity (DM), length of panicle (PL), weight of panicle (WP), spikelet per panicle (SPP), test weight (TW), grain yield per plant (YPP), and four qualitative traits *viz.*, length/breadth ratio before cooking (L/B Ratio BC), length/breadth ratio after cooking (L/B Ratio AC), amylose content (AC) and gel consistency (GC).

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Statistical analysis

Adequacy of scale should fulfil two conditions namely, additivity of gene effects and independence of heritable components from non-heritable ones. The test of first condition gives information about absence or presence of gene interactions. The test of adequacy of scales is essential because in most of the cases the estimation of additive and dominance components of variances is made assuming the absence of gene interaction. The generation mean analysis was performed according to [6] and [8] for the estimation of genetic components of variation, epistasis model and gene effects in two steps (i) testing for epistasis to determine the presence or absence of interallelic interaction and (ii) estimation of gene effects, variances and the type of epistasis involved. Scaling test for A, B, C and D scales as suggested by [7] and [12] was applied to test the adequacy of simple additive-dominance model but since, back cross is absent in the present study, I have considered only C & D scale and it is computed as follows:

$$C = 4\overline{F}_2 - 2\overline{F}_1 - \overline{P}_1 - \overline{P}_2$$

$$D = 4\overline{F}_3 - 2\overline{F}_2 - \overline{P}_1 - \overline{P}_2$$

When the scale is adequate, the values of A,B, C and D should be zero within the limit of their respective Standard Errors.

Variances of above scales:

 $\begin{array}{c} \mathsf{V}_{\mathsf{C}} = \mathsf{16V}(\overline{F}_2) + \mathsf{4V}(\overline{F}_1) + \mathsf{V}(\overline{P}_1) + \mathsf{V}(\overline{P}_2) \\ \mathsf{V}_{\mathsf{D}} = \mathsf{16V}(\overline{F}_3) + \mathsf{4V}(\overline{F}_2) + \mathsf{V}(\overline{P}_1) + \mathsf{V}(\overline{P}_2) \end{array}$ Standard errors of the above scale: $\begin{array}{c} \mathrm{SEc} = & 2\sqrt{Vc} \\ \mathrm{SEd} = & 2\sqrt{Vd} \\ \mathrm{Now, \ the \ 't' \ values \ are \ calculated \ as \ follows:} \\ \mathrm{tC} = \mathsf{C}/\mathsf{SEC} \\ \mathrm{tD} = \mathsf{C}/\mathsf{SED} \end{array}$

The calculated value of 't' are to be compared with tabulated value of 't' at 5% level of significance. In each test, the degree of freedom is sum of the degrees of freedom of various generations (total number of observations - total number of replications) involved. However, in case of un-replicated data, the degrees of freedom will be number of observations.

RESULTS AND DISCUSSION

Mean performance of traits in different generations

To detect the relative importance of the components of genetic variation, additive (d), dominance (h) and epistatic contributions additive × additive (i) and dominance × dominance (I) were estimated by portioning the population means of parents, F1 and F2. The estimates of the above components based on the five parameter model using the population mean are presented character wise in table 1. Interacting and non-interacting traits were sorted out with the help of scaling test [14]. The scales deviating from zero indicates the presence of non-allelic interaction, which showed inadequacy of additive-dominance model. In such condition, five parameter model was used to estimate the gene effects (m,d and h) and their interactions (i and I) following [6] and [8]. Progenies of the cross between Swarna sub-1× Tetep were advanced to F2, and F3to isolate high yielding segregants with sheath blight resistance. To elucidate the nature of gene action for yield traits and sheath blight resistance, generation mean analysis was carried out using the data recorded from five generations of the above cross combination. The character wise mean performances of the five generation materials P₁, P₂, F₁, F₂, and F₃ for 14 traits were presented in [Table-2]. F₁ along with two segregating populations (F₂ and F₃) flowered and matured earlier compared to parents, which was desirable in further selections. Plant height of F₁s were mostly like donor parent (Tetep) while segregating population F2andF3had mostly intermediate to slightly higher plant height than both of the parents. These findings are in agreement with earlier report by [22,27] for plant height. The number of reproductive tillers per plant in F1 and all segregating populations (F2 and F3) was intermediate to the parents. The less difference for panicle length and weight was observed between parents. Among all generation materials panicle length and weight was comparable to both parents. The F2 segregants possessed higher panicle length whereas weight of panicle was higher in F₃population. The F₂ and F₃ segregants possessed less total number of spikelet per panicle than F1 and recurrent population. The total number of spikelet per panicle in F1 was higher than

both parents and both segregating populations (F₂ and F₃). The test weight of all generation materials was intermediate to the parents, and desirable for consumer preference. The kernel length and breadth ratio before and after cooking was higher in F_1 than both parents while intermediate in F_2 and F_3 segregating generations. Amylose content was higher in F1 and both segregating population than the recurrent parent while in segregation populations, F₃ had lower amylose content than the F_1 and F_2 populations. Gel consistency was intermediate in F_1 and segregating populations than both of the parents but within generation materials, F₃ had higher GC than F₁ and F₂. Most of the above results of present investigation are conformity with the findings of [22,26,27]. The Swarna sub-1 recorded higher grain yield per plant compared with Tetep while the F1 yielded more grain yield compared with the donor parent but less than the recurrent parent, but in the two segregating populations (F2 and F3), grain yield per plant were intermediate than non-segregating generations. The range of percent disease severity was presented in Fig. 1 and reaction to sheath blight disease of the parents shown in Plate 1. The sheath blight susceptible parent Swarna sub-1 showed high disease severity (60.46%) compared with resistant parent Tetep (17.72%) while intermediate disease severity was observed in F_1 and three segregating populations. Among F₁, F₂ and F₃ population, F₁ showed less disease severity (20.09%) than F₂ and F₃ populations. Singh et al., [28] reported that while transferring sheath blight resistance QTLs, notably, one line (Pusa 1604-05-3-5) possessing single QTLqSBR11-1 showed comparable disease reaction score as that of the donor parent, Tetep while Pusa1604-05- 45-1 possessing two and Pusa1604-05-43-1 with three sheath blight resistance QTLs, showed moderate resistance only. This could be attributed to several reasons such as QTL-marker recombination [15], QTL-background interactions [31,2], presence of un-known QTLs in the improved lines derived from donor [30] and/due to the competitive effectiveness of qSBR11-1 than the other two QTLs [17]. Further, disease resistance variations among the lines positive for all QTLs, can be attributed to epistatic effects and QTL-background interaction, a well-recognized component of natural genetic variation [11].

Estimates from scaling tests

Scaling tests were performed to understand the adequacy of simple additivedominance model (Table 2). The scaling test showed all C and D scales were significant for plant height, days to maturity, panicle length, weight of panicle, number of spiklets per panicle, test weight, yield per plant, length/breadth ratio before cooking, length/breadth ratio after cooking, gel consistency and percentage of disease severity indicating presence of epistasis. All the traits related to yield as well as sheath blight resistance in the present study were significant in either one of the scales or in combination representing the existence of epistatic interactions between the genes involved except in case of panicle weight with none of the scales showing significance as observed earlier by Singh *et al.*, [27] for all the traits studied and reported model was sufficient.

Estimation of gene effects based on five generation means

Digenic non-allelic interaction model with five parameters namely m, d, h, I and I revealed that the epistatic interaction model was found adequate to explain the gene action in the traits number of reproductive tillers, plant height, days to maturity, days to 50% flowering, panicle length, weight of panicle, total number of spikelet per panicle, test weight, yield per plant, length and breadth ratio before cooking, length and breadth ratio after cooking, amylose content, gel consistency and disease severity. The estimates of gene effect clearly illustrate high variation in the observed traits (Table 2). Mean and additive components for number of reproductive tillers, Plant height, days to maturity, days to 50% flowering, panicle length, weight of panicle, total number of spikelet per panicle, test weight, yield per plant, length and breadth ratio before cooking, length and breadth ratio after cooking, amylose content, gel consistency and disease severity were highly significant. The dominance (h) and dominance × dominance (l) gene effects displayed opposite signs for the traits viz., number of reproductive tillers, plant height, days to maturity, lengthand breadth ratio after cooking and gel consistency indicating duplicate epistasis. Most of these results are in conformity with the earlier reports of Divya et al., ([3] for plant height, number of productive tillers,

panicle length, days to Maturity, length and breadth ratio after cooking and gel consistency. On the contrary, Singh *et al.*, [27] reported same sign for the trait days to maturity indicating complementary recessive epistasis. The values of dominance (h) and dominance \times dominance (l) interaction were in the same direction for traits like days to 50% flowering, panicle length, weight of panicle, number of spikelets per panicle, test weight, yield per plant, length and breadth ratio before cooking, amylose content and disease severity and the interaction fit into complementary epistasis model. It was reported that gene effects are known to be cross specific and fits into complementary recessive epistasis for grain yield [29].On the contrary, Singh *et al.*, [27] reported opposite sign for the traits like days to 50% flowering, weight of panicle, number of spikelets per panicle, test weight, yield per plant and disease severity indicating duplicate epistasis.

The classification of gene interactions depends on the magnitudes and signs of the estimates of dominance and dominance × dominance effects, when there are many pairs of interacting genes [13]. The sign associated with the estimates of (d) and (h) indicates the parent that concentrates the highest number of genes for increasing the trait [5]. Additive effect was the only significant portion of gene controlling grain yield per plant of the rice. Finally, additive and dominance gene effects were found important in controlling sheath blight disease reaction. The plus sign in the additive gene effect implies that Swarna sub-1 contributes positively to the trait as compared to Tetep, and vice versa. The positive sign for (d) was observed in the traits days to 50% flowering, panicle length, weight of panicle, number of spikelets per panicle, test weight, yield per plant, length and breadth ratio before cooking, amylose content and disease severity, while the negative sign for (h) was observed in the traits number of reproductive tillers, plant height, days to maturity, length and breadth ratio after cooking and gel consistency demonstrated that the dominance was towards the resistant parent Tetep as observed earlier [29,21,25] which explained additive effect in yield and disease related traits in rice. On the contrary, Paul et al., [18]; Li et al., [9]; Singh et al., [27] have reported the importance of dominance genetic effects.

Conclusion

The generation mean for most of the characters showed the importance of both additive and dominance type of gene effects. However, additive effect was the only significant portion of gene controlling grain yield per plant, while additive and dominance gene effects were found important in controlling sheath blight disease reaction. Among the epistatic gene effects, the additive genetic variance was predominant in case of days to maturity and number of spikelets per panicle, and it is associated with homozygosity and hence it is fixable in nature and selection for these traits will be very effective whereas rest of the traits shown predominant of dominant genetic variance and hence it is not fixable and selection for these traits will be postponed to later generations until homozygosity is achieved.

Application of research: The importance of this experiment is imparting information on genetics of various contributing traits of resistance and yield related which would further help in choosing appropriate breeding strategy for sheath blight resistance and yield enhancement in rice.

Research Category: Genetics and Plant Breeding

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