

Original Research Article

Genetic analysis of quantitative traits, gall midge and brown spot tolerance in rice (*Oryza sativa* L.) genotypes under irrigated ecosystem

ABSTRACT

Gall midge and brown spot of rice are a fungal disease that affect both seedlings and mature plants. To investigate resistance of rice to this disease twenty nine rice cultures were evaluated under irrigated ecosystem at Regional Agricultural Research Station, Jagtial during *kharif*, 2020. High heritable estimates were observed for all the 8 traits studied. High estimates of heritability was recorded for important yield attributes viz., days to 50% flowering (91.4, 11.24), plant height (77.8, 9.53), 1000-grain weight (98.4, 54.5), number of grains per panicle (78.3, 40.18) and grain yield (98.7, 33.87) indicated that these traits were under the control of additive genes. Significantly positive correlations were recorded with plant height, Number of productive tillers per plant and 1000 seed weight at both genotypic and phenotypic levels, the results clearly indicated that long duration genotypes with more height and bolder grains contribute for more grain yield. Among the eight traits studied, 1000 grain weight contributed highest (67.8%) towards total divergence followed by days to 50% flowering (11.5%), number of grains per panicle (6.06) and Galls midge incidence percentage (6.06%). Genotypes, JGL 38957 of cluster II, JGL 38950 of cluster III, JGL 38935 of cluster IV and JGL 38921 of Cluster VII could be the best source to develop brown spot, gall midge resistance and high yielding rice varieties.

Key words: Brown spot, correlation, gall midge, genetic diversity, heritability and rice.

Introduction:

“Rice (*Oryza sativa* L.) is one of the most important food crops of the world feeding more than half of the global population. India ranks second in production and consumption of rice next to China. Genetic manipulation is one of the best means for enhancing the yield levels of rice to cope the increasing global population. The success of breeding programme lies on the fact that the parents involved in any particular cross should be genetically divergent” (Deniel 2000). “The germplasm provides immense scope for wide variability. Among biotic stresses, brown spot caused by *Bipolaris oryzae* is one of the most important fungal diseases of rice in irrigated rice ecosystem causing as high as 45% yield reduction in severe epidemics” (IRRI, 2012). “This disease under favorable conditions becomes hazardous to rice crop. Brown Spot is currently regarded as a serious rice disease worldwide” (Barnwal *et al.*, 2013; Mizobuchi *et al.*, 2016). “This pathogen causes quantity and quality losses that are associated with the disease incidence on the leaves and grains” (Ou, 1985; Lee, 1992).

“Another important biotic stress factor causing huge yield losses is gall midge caused by *Orseolia oryzae*. The incidence of gall midge is severe in delayed planting conditions, but the recent observations of this pest incidence across the country revealed that, the incidence also increased even under normal planting situations. The Asian rice gall midge [*Orseolia oryzae*

(Wood-Mason)] is an important rice pest causing an annual average yield loss of about US \$80 million in India” (Nidhi *et al.*, 2012). “Breeding for the refinement of rice varieties with high yields along with gall midge tolerance is essential and should be continuous due to rapid and continuous evolution of new biotypes of gall midge. Knowledge on the heritability of genetic trait is essential to the plant breeders in determining the response to selection and to provide the information on the extent of transmissibility of that selected trait of interest to the progenies in the subsequent generations” (Sabesan *et al.*, 2009). In addition, high genetic advance coupled with high heritability helps breeders in accurate calculation of the genetic gain under selection. The relationship of these traits *viz.*, gall midge and brown spot tolerance with yield and yield attributing traits would be helpful in selecting high yielding genotypes, coupled with gallmidge or brown spot tolerance.

Keeping in view of the above, the objectives were framed to study the variability, heritability, genetic advance and identify the genotypes with gall midge and brown spot tolerance to use them in further breeding programmes.

Materials and Methods:

The material for the present study consists of 33 rice genotypes developed through pedigree method of breeding and evaluated at Regional Agricultural Research Station, Polasa, Jagtial, during *khariif*, 2020. Thirty days old seedlings of each entry were transplanted in a Randomized Block Design with two replications of plot size of 8.28m². Data on plant height (cm), panicle length (cm), number of grains per panicle and number of productive tillers (per m² area) was recorded on 5 random plants for each replication and each entry at the time of maturity. However, days to 50 % flowering and grain yield (kg per ha.) were recorded on whole plot basis, whereas, random sample was taken to estimate 1000 grain weight (g) for each entry in each replication. The incidence of gall midge was recorded as percent tillers affected with silver shoots (SES, IRRI, 2013) on 10 random plants and averaged. The symptoms of brown spot disease score was taken for each entry from all the three (3) replications and then averaged (Table 4 and Table 5). Scoring of disease incidence (0 to 9 scale) was done based on the affected leaf area as per standard Evaluation system (SES), IRRI 2013. The mean data after computing for each trait was subjected to statistical analysis *viz.*, analysis of variance, genotypic coefficient of variation (GCV), phenotypic coefficients of variation (PCV) (Sivasubramanian and Madhavamenon, 1973), heritability (h²) in the broad sense (Singh and Chaudhary, 1985), genetic advance and correlations (Johnson *et al.*, 1955). The genetic divergence for traits studied in the present experimental material was estimated using Mahalanobis (1936) D² statistics. “Allotment of rice cultures into different clusters was carried out following Tocher’s method” (Rao, 1952).

List 1 : Brown spot scoring as per SES, IRRI (2013)

Score	Severity: % leaf area diseased
0	No disease observed
1	Less than 1%
2	1-3%
3	4-5%
4	6-10%
5	11-15%
6	16-25%
7	26-50%
8	51-75%
9	76-90%

List 2 : Gall midge scoring as per SES, IRRI, 2013

Score	Infected tillers in field test
0	No injury
1	Less than 1%
3	1-5%
5	6-10%
7	11-25%
9	More than 25%

Results and Discussion:

Phenotypic and genotypic coefficient of variation presented in **Table 1** indicated the presence of good amount of variability for the 8 traits studied in 33 genotypes. This variability was studied as heritable and non heritable variation. Days to 50% flowering, effective bearing tillers/m,² plant height and panicle length exhibited low levels of PCV and GCV values, therefore for improvement of these characters in desirable direction, creation of variability through hybridization followed by selection in segregating generations is the best practice, whereas, the important yield attributes viz., 1000 grain weight and number of grains/panicles recorded moderate values. Hence simple selections based on phenotypic superiority would improve these traits. These results are in agreement with the findings obtained by Sameera *et al.*,(2015), Srinivas *et al.*,(2016) , Ajmera *et al.*,(2017) and Saha *et al.*,(2019) for number of grains per panicle; Ahmed *et al.*,(2010), Sameera *et al.*,(2015), Ajmera *et al.*,(2017) and Saha *et al.*,(2019) for 1000-grain weight; Ajmera *et al.*,(2017) and Saha *et al.*,(2019) for number of productive tillers per plant; Mohan *et al.*,(2015) and Thippaswamy *et al.*,(2016) for gall midge incidence; Allam *et al.*,(2015), Bhati *et al.*,(2015), Ajmera *et al.*,(2017), Behera *et al.*,(2018) and Saha *et al.*,(2019) for grain yield. The dependent trait grain yield exhibited moderate values for both PCV and GCV, Rice workers, Akinwale *et al.*,(2011) and Ramanjaneyulu *et al.*,(2014) reported moderate GCV and high PCV values for grain yield which are in accordance with present findings. Highest values for PCV than GCV for all the traits could be due to greater interaction of environment factors like climate, soil *etc.* with genotypes therefore there could be a major role of environment in expression of these traits and consideration of environment into account is more practical while selecting for the characters in positive direction. Similar finding were reported by Vanisree *et al.*,(2013), Ketan and Sarkar (2014), Mohan *et al.*,(2015), Srinivas *et al.*,(2016), Ajmera *et al.*,(2017) and Gyawali *et al.*,(2018).

It was observed that PCV was slightly higher than GCV for days to 50% flowering, plant height, panicle length and 1000 seed weight reflecting less influence of environment in the expression of traits and greater role of genetic control governing the characters is in agreement with the results explained by Karim *et al.*,(2007), Sravan *et al.*,(2012), Mohan *et al.*,(2015),

Ajmera *et al.*,(2017), Behera *et al.*,(2018) and Saha *et al.*,(2019). However, estimates of PCV were considerably higher than GCV for number of productive tillers per m², number of grains per panicle, gall midge incidence and grain yield indicating the sensitive nature of these traits to environmental fluctuations and predominance of non-additive gene effects. Higher estimates of PCV than GCV for per cent incidence of galls reflects the major role of environment in that particular season of experimental evaluation in inducing the pest incidence among the genotypes. Similar findings are in accordance with the earlier reports of Mohan *et al.*, (2015), Thippaswamy *et al.*,(2016) and Adhikari *et al.*,(2018) for effective bearing tillers, number of grains per panicle and grain yield.

Heritability measures the relative degree of transmission of a character from parents to their off springs. The quantity of heritable variation of a character transmitted from parents to offspring is the key for improvement of that trait. High estimates of heritability was recorded for days to 50% flowering, plant height, 1000-grain weight, number of grains per panicle and grain yield (Table 1), which implies that these traits are genetically unchanged as they are less influenced by environment and these traits were improved by mere selection. Whereas, number of productive tillers per m², panicle length and gall midge incidence had relatively moderate estimates; hence, improvement through selection could be low due to masking effect of environment on the expression of these traits (Table 1). Therefore, while selecting the genotypes showing resistance to gall midge, a breeder should completely aware of the favorable conditions that enhance the pest incidence and ensure that there should be sufficient pest load in that particular season of evaluation. Similarly, moderate heritable values were reported by Sangram kumar *et al.*, (2011), Thomas and Gabriel (2012) for test weight, Ramanjaneyulu *et al.*,(2014) for days to 50% flowering and panicle length. The genotypic coefficient of variance (GCV) and phenotypic coefficient of variance (PCV) were high for gall midge incidence (43.15, 67.98), here PCV were considerably high than the GCV estimates indicating the existence of environmental role in expression and predominance of non-additive gene effect.

Table 1: Genetic parameters for Gall midge incidence, yield and its contributing traits in rice

Character	Mean	Range		GCV	PCV	h ² (Broad sense)	Genetic advance in % over mean (at 5%)
		Min	Max				
Days to 50% flowering	99.0	92	110	5.7	5.9	91.4	11.2
Effective bearing tillers per m ²	381.2	304	425	5.7	8.6	45.0	7.9
Plant height (cm)	112.5	95	123	5.2	5.9	77.8	9.5
Panicle length (cm)	24.5	22	27	3.2	4.9	42.0	4.2
1000 grain weight (g)	21.8	12	30	26.6	26.8	98.4	54.5
Number of grains per panicle	178.0	102	282	22.0	24.9	78.3	40.1
Seed yield (kg/ha)	4609.8	2320	6265	19.8	23.9	68.7	33.8
Galls midge incidence (percent tillers)	6.8	1.3	14.8	43.1	67.9	40.3	56.4

“Genetic advance is the genetic gain in selected individuals over parental population resulting from the application of selection pressure and the values facilitate the breeder to design appropriate breeding programme for improvement. Heritability values along with the estimates of genetic advance should jointly be considered to arrive at a more reliable conclusion” (Johnson *et al.*, 1955). In the present investigation, 1000 seed weight, number of grains per panicle and seed yield showed high heritability combined with high genetic advance values reflecting the existence of additive gene action in the expression of these traits and hence selection would be effective as investigated” by Karande *et al.*,(2015), Ajmera *et al.*,(2017) and Saha *et al.*,(2019) for number of grains per panicle; Toshimenla and Changkija (2013), Chandramohan *et al.*, (2016), Islam *et al.*,(2016), Srinivas *et al.*,(2016) and Ajmera *et al.*,(2017) for 1000- grain weight; Mohan *et al.*,(2015) for gall midge incidence; Rahman *et al.*,(2014) , Karande *et al.*,(2015) and Ajmera *et al.*,(2017) for grain yield. Similarly, moderate to high values of heritability and genetic advance were observed for galls midge incidence revealed the predominance of additive gene effects and simple selection would be rewarding for improvement of the traits. High heritability coupled with moderate genetic advance was recorded for days to 50% flowering. Ketan and Sarkar (2014), Chandramohan *et al.*,(2016) and Behera *et al.*,(2018) found the “same result for panicle length suggesting the role of both additive and non additive gene effects in their inheritance, therefore, adoption of breeding procedures which could exploit both the gene actions”. “Greater values for three genetic components *viz.*, GCV, heritability and genetic advance specify the action of additive genes that control the expression of characters. Important yield contributing characters *viz.*, 1000 grain weight and number of grains per panicle showed higher values for these three genetic components indicating the role of additive gene controlling these traits, thus these characters can be improved by practicing simple selection breeding method. Yield being a complex and dependent trait also exhibited higher values for these genetic components” [SRINIVAS *et al.*2021].

Character association studies between yield and other traits revealed that, among the eight traits studied for 33 genotypes, high magnitude of genotypic correlation coefficients values (plant height and number of grains per panicle) were observed in most of the cases as compared with the corresponding phenotypic correlation coefficients, indicating the negligible influence of environment factors (Table 2). Similar results were reported by Bhattacharya *et al.*,(2007) Ravindra Babu *et al.*,(2012), Hossain *et al.*,(2015), Mohan *et al.*,(2015), Ratna *et al.*,(2015) and Kalyan *et al.*,(2017). In some cases, phenotypic correlation coefficients were higher than their genotypic correlation coefficients (effective bearing tillers per m², panicle length and test weight), which indicate the suppressing effect of the environment that can alter the expression of the characters at the phenotypic level. Rice yield recorded significantly positive correlation with plant height, number of productive tillers per plant and 1000 seed weight at both genotypic and phenotypic levels, the results clearly indicated that long duration genotypes with more height and bolder grains contribute for more grain yield. Test weight had positive correlations with gall midge incidence indicating fine grain varieties were relatively tolerant and bold grain varieties were susceptible. The results are in agreement with Saha *et al.*,(2019) for days to 50% flowering

and number of grains per panicle. Similarly, positive correlation of panicle length, 1000-grain weight and straw yield on grain yield was also reported by Kumar *et al.*, (2018). However, negative relationship between days to 50% flowering and grain yield was reported by Babu *et al.*, (2006) and Chandan Kumar and Nilanjaya (2014). Whereas, grain yield was not associated significantly with gall midge incidence (Table 2). Similar findings were reported by Bashir *et al.*, (2013) for grain yield.

“Mating between the parents having more divergence, generate high yielding potential offspring’s rather than less divergent parents. The degree of divergence among the genotypes for a given set of traits can be explained by grouping them into different clusters. Phenotypic divergence does not reflect the actual divergence. Crosses between phenotypically divergent parents may not generate elite progenies” [SRINIVAS *et al.* 2021]. In the present investigation, 33 genotypes were grouped into 11 clusters (Table 3) with highest number of genotypes (13) allotted to cluster I followed by cluster V (6 genotypes), cluster III (5 genotypes) and cluster XI (2 genotypes). Remaining clusters *viz.*, II, IV, VI, VII, VIII, IX and X were accommodated with single genotype each. Higher the distance between the clusters, more is the divergence between them. The highest distance (352.6) was observed between clusters V and VIII (Table 3) followed by VIII and IX (345.01), V and X (311.50), VI and IX (303.24), IV and V (294.61) and V and VI (292.13). Hence parents from these divergent clusters can be used in crossing programme for developing high heterotic hybrids and thereby chance of obtaining desirable transgressive segregants in succeeding generations. Cluster means (Table 4) provides useful information for breeders to choose suitable parents based on character means of respective clusters. In the present experimental material, the trait 1000 grain weight contributed highest (67.8%) towards total divergence (Table 4) followed by days to 50% flowering (11.5%), number of grains per panicle (6.06) and gall midge incidence percentage (6.06%), whereas, least contribution towards divergence was observed for panicle length (0.37%) and effective bearing tillers/m² (0.95%). Thus, development of varieties with different grain segments like coarse, fine and super fine and different maturity groups could be possible.

The genotype (JGL 38935) in cluster IV was recorded highest test weight and high grain yield, low percent incidence in gall midge and brown spot whereas, the genotype (JGL 38900) in cluster III had low test weight and was found to be susceptible to gall midge and brown spot. Hybridization program should always be formulated in such a way that the parents belonging to different clusters with maximum divergence to get desirable transgressive segregants. Based on the inter cluster distances and means of cluster groups, the present investigation suggests that the genotypes grouped in cluster I could be crossed with JGL 38944 of cluster IX with a view to derive transgressive segregants for improvement of grain yield, test weight and tolerant to gall midge and brown spot. The genotypes JGL 38950 of cluster III and JGL 38935 of cluster IV having least gall midge incidence (1.3%) and brown spot score (zero) respectively, could be utilized in hybridization program for development of gall midge and brown spot resistant rice varieties.

“Brown spot incidence is observed mostly in drought and high temperature conditions, causing severe yield losses. The optimum temperature for growth and conidial germination has been found to be 27-30°C and 25-30°C, respectively” (Ou, 1985). “Many efforts have been made towards searching for resistance to brown spot” (Chakrabarti, 2001; Nagai and Hara, 1930). The present investigation is aimed at the identification of brown spot resistant lines under field screening and incidence was measured in 0 to 9 score as per SES, 2013. Genotypes exhibited greater variation for the incidence from highly susceptible to resistant. Four genotypes *i.e.*, JGL 38957, JGL 38955, JGL 38935 and JGL 38917 recorded resistant reaction for brown spot and less than 5% of gall midge incidence, while, four genotypes *i.e.*, JGL 38953, JGL 38935, JGL 38934 and JGL 38921 exhibited resistant to moderate reaction as they recorded least damage score of 3 and less than 10% of gall midge incidence with good yields, whereas, remaining genotypes showed moderate to highly susceptible reaction. Satija *et al.*, (2005) identified “15 resistant rice cultivars which recorded less than 5% damage. Two genotypes *i.e.*, JGL 38957 and JGL 38921 could be identified as the best source to develop brown spot, gall midge resistance and high yielding varieties as these entries fall under cluster II and Cluster VII respectively, having high means for yield and important yield attributes like effective bearing tillers/m², panicle length and 1000 seed weight. Breeders can make best use of JGL38957 exhibiting resistant reaction for gall midge and brown spot with short duration in nature to generate high yielding resistant varieties”.

Summary and conclusions:

In the current examination of rice genetic analysis, larger PCV values than GCV values were found for every yield, with the attributed features demonstrating strong heritability estimates. The important yield attributing traits *viz.*, 1000 grain weight and number of grains per panicle recorded both high heritability and genetic advance values. Yield attributing traits like 1000 seed weight, effective bearing tillers per m² exhibited significant and positive association with yield. Genotypes from cluster II, III, IV and VI could be used in crossing programme for development of high yielding varieties resistant to gall midge and brown spot and the genotypes from cluster IV and VIII could be used in breeding programme to develop high yield and brown spot resistant varieties. Six genotypes *viz.*, JGL 38953, JGL 38955, JGL 38935, JGL 38917, JGL 38921 and JGL 38957 were identified as good sources for gall midge, brown spot resistance and high yielding ability. These six genotypes could be released as new varieties and used as donors for future breeding programmes.

Table 4: Cluster mean for biotic stress, yield and its attributing traits

Cluster Means : Tocher Method									
	Days to 50% flowering	Effective bearing tillers/m²	Plant height(cm)	Panicle length (Cm)	1000 grain weight (g)	Number of grains/panicle	Yield (kg/ha)	Galls midge incidence (percent tillers)	Brown spot
Cluster 1	94.69	381.62	114.48	25.01	26.02	157.15	5172.69	7.35	2
Cluster 2	92.50	420.20	117.10	24.80	22.60	217.00	4634.00	7.58	2
Cluster 3	100.00	367.84	111.14	25.58	16.13	218.30	3261.00	8.75	3
Cluster 4	99.00	420.20	114.40	23.70	29.35	150.50	5586.00	2.40	0
Cluster 5	106.75	392.70	114.62	23.85	14.28	210.00	4223.25	4.98	4
Cluster 6	102.50	352.00	105.05	24.40	29.33	150.00	5272.50	3.50	1
Cluster 7	92.50	303.60	97.20	24.20	16.91	222.50	3220.50	5.15	4
Cluster 8	102.50	389.40	104.00	24.40	30.16	139.00	6126.00	14.20	1
Cluster 9	96.50	338.80	108.70	24.40	13.00	124.50	3814.50	7.63	2
Cluster 10	96.00	389.40	94.70	22.80	25.86	102.00	4087.00	1.58	3
Cluster 11	108.50	404.80	120.20	22.80	23.82	188.25	5246.75	6.99	5
%Contribution of character	11.5	0.95	5.3	0.37	67.8	6.06	1.89	6.06	-

Table 5: Distribution of genotypes to different clusters based on Tochers method

Cluster Number	S. No.	Genotypes	Pedigree	Yield (t/ha)	Galls midge incidence (percent tillers)	GM score	Brown spot score
I	24	JGL 38952	JGL 25945 XJGL 24423	5220	6.1	5	4
	28	JGL 38956	JGL 25945 XJGL 24423	5099	4.4	3	2
	23	JGL 38951	JGL 25945 XJGL 24423	4542	9.5	5	5
	29	JGL 38957	JGL 25945 XJGL 24423	6265	7.1	5	1
	26	JGL 38954	JGL 25945 XJGL 24423	4887	11.9	7	2
	25	JGL 38953	JGL 25945 XJGL 24423	5827	12.6	7	1
	27	JGL 38955	JGL 25945 XJGL 24423	5894	2.2	3	2
	14	JGL 38927	JGL 18047 X GSR29	4768	10.3	5	3
	9	JGL 38915	WDR 359 X JGL 18047	4799	8.0	5	1
	13	JGL 38924	JMS 19B X GP228	4538	6.1	5	2
	10	JGL 38917	JMS 11B X CMS64B	5327	2.9	3	1
	17	JGL 38937	JGL 25960 X IRTON 270	4499	3.2	3	1
	12	JGL 38922	WDR 359 X CR 1898-32-62-CN-12-2	5583	11.3	7	1
II	31	JGL 18047		4634	7.6	5	2
III	1	JGL 38900	WGL 14 X GP227	3231	14.8	7	3
	3	JGL 38902	WGL 14 X GP227	3449	7.3	5	2
	2	JGL38901	WGL 14 X GP227	3196	8.6	5	4
	21	JGL 38945	JGL 135 X JGL 28921	4110	11.8	7	1
	22	JGL 38950	JGL 28921 X NLR3042	2320	1.3	3	5
IV	16	JGL 38935	JGL 25960 X IRTON 270	5586	2.4	3	0
V	4	JGL 38903	WGL 23985 X GP 270	4977	2.8	3	4
	32	JGL 11470		4714	7.1	5	6
	30	RNR15048		3115	11.5	7	3
	5	JGL 38904	WGL 23985 X GP 270	4415	3.6	3	4
	19	JGL 38941	JGL 135 X JGL 28921	4978	2.9	3	1
	7	JGL 38908	NLR 4001 X JGL 13595	3142	2.1	3	7
VI	15	JGL 38934	JGL 25960 X IRTON 270	5273	3.5	3	1
VII	8	JGL 38909	NLR 4001 X JGL 13595	3221	5.2	3	4
VIII	11	JGL 38921	WDR 359 X CR 1898-32-62-CN-12-2	6126	14.2	7	1
IX	20	JGL 38944	JGL 135 X JGL 28921	3815	7.6	5	2
X	18	JGL 38938	JGL 25960 X IRTON 270	4087	1.6	3	3

XI	6	JGL 38906	JGL 11470 XGSR27	4254	6.6	5	6
	33	MTU 1001		6240	7.4	5	4

UNDER PEER REVIEW

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