

Genetic Analysis of New Plant Type in Rice (*Oryza sativa* L.) with a View to Exploit Yield Potential

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ABSTRACT. *The genetical architectures of twelve quantitative characters of interest have been investigated using the basic generations and F₃ families from two crosses, chosen at random from IRRI's New Plant Type (NPT) programme (*japonica* × *javanica*) with the objective of increasing the genetic yield potential to 13-15 t/ha of direct seeded, irrigated crop in lowland tropics. The experimental plants were raised in a completely randomized experiment for each cross. The parents of both crosses differed significantly for most of the characters under study, except for proportion of filled spikelets (PFS) and grain yield (GY) in Cross 1, and grain weight (GW) in Cross 2. Only three of these characters, panicle number (PN), GY and dry matter (DM), in Cross 1 displayed heterosis. The genes controlling the characters in Cross 1 displayed mainly additive or additive and dominance effects, except for two related characters, tiller number (TN), and PN, which displayed 'j' type epistasis. In contrast, those controlling all but two of the characters in the second cross displayed epistasis, which for most was of the duplicate type. The results show that characters in both crosses were heritable except for harvest index (HI) in Cross 1 and heritabilities varied from low to high. Genetic correlations between characters in Cross 1 were very different from those in Cross 2, which suggest that the chief cause of these correlations is the linkage disequilibrium of linked genes, rather than pleiotropy. An assessment of the potential of these crosses showed that, while it should be relatively easy to achieve the NPT targets for six characters in both crosses, this is unlikely for PFS, GY and HI with one cycle of inbreeding.*

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INTRODUCTION

The belief among rice breeders that tropical cultivars of the *indica* type have reached a yield plateau of 10 t/ha, has caused the breeders at International Rice Research Institute (IRRI) to switch their programme to the breeding of a radically different ideotype, known as New Plant Type (NPT) for direct seeded, irrigated rice crop which would yield 13-15 t/ha, in lowland tropics. The chief characteristics of the NPT, were determined considering several different perspectives (Vergara, 1988; Janoria, 1989; Dingkuhn *et al.*, 1991), such that the plant should produce 3-4 tillers, all of which produce panicles containing 200-250 grains on very sturdy stems, should bear dark green leaves, should be about 90 cm in height and 100-130 days growth duration and should have multiple pest and disease resistance and a harvest index of 0.6. Some of these characteristics lie well outside the normal range of the variation found in *indica* cultivars which in any case, are believed to have insufficient genetic variation for yield. Therefore the NPT breeding programme has been initiated from the crosses between *japonica* and *javanica* (according to Glaszmann (1987), refers to as tropical *japonica*) varieties (IRRI, 1989). However knowledge on genetical architecture of the characters of interest in the NPT programme is not well known. This paper reports a genetic analysis of two of the *japonica* × *javanica* crosses chosen at random from the IRRI's NPT programme.

MATERIALS AND METHODS

Two of the crosses chosen for the investigation were the Jinmiby eo × Gaok (Cross 1) and Sangnambatbyeo × Kemandi Pance (Cross 2). Jinmiby eo and Sangnambatbyeo are the two Korean varieties that belong to *japonica* type. These female varieties were chosen because of their short stature, high yielding ability under temperate conditions, dense spikelets per panicle and cross-compatibility with *javanica* types. Male parents Gaok and Kemandi Pance are two traditional Bulu (*Javanica*) varieties from Indonesia that were chosen because of their large panicles, low number of tillers, sturdy stems and dark green and thick leaves.

Three groups of families namely basic generations (P_1 , P_2 , F_1 , F_2 , BC_1 , BC_2), F_3 families and back crosses to F_2 individuals ($L_1 = F_2 \times P_1$, $L_2 = F_2 \times P_2$, $L_3 = F_2 \times F_1$) of the same age were raised at IRRI, Phillipines in the dry season of 1994, in two adjacent blocks in a completely randomized design, one block per cross. In each cross, 20 plants were raised from each of the non-segregating families, 56 from F_2 population, 50 from each of the first back

crosses and 30 F_3 families with 8 plants from each family. Eight plants were also raised in each of $35 \times 3F_2$ back cross families for Cross 1 and $32 \times 3F_2$ such families for Cross 2. Two recently improved IRRI varieties, IR72 and IR74 were also included in the two experiments. This paper reports the genetic analysis based on basic generations and F_3 families.

All of the cross seeds were produced using modified vacuum (Herera and Coffman, 1974) emasculation method followed by hand pollination the following day. The F_3 families, IR72 and IR74 in the experiment were from naturally self pollinated seeds. All the experimental seeds were pre-germinated in petridishes and were sown in $5 \times 5 \times 5$ cm small plastic pots filled with sterilized soil, one seed being sown in each pot. The pots of each experiment, one for each cross were arranged in a pair of completely randomized block designs (CRBD) in the green house. Experiment 1 (Cross 1) consisted of 1336 individually randomized plants and experiment 2 (Cross 2) with 1264 plants. After three weeks, plants were transplanted in the field in the same CRBD at a spacing of 30×20 cm at IRRI, Philippines in the dry season of 1994. The experiments were managed in the usual way except for the special efforts made to keep them free from pests and diseases as much as possible. Each of the plants included in the experiment 1 and experiment 2 were scored for the following twelve characters:

- Days to heading (DH) - Number of days from the date of sowing to the first panicle emerged from the flag leaf sheath;
- Days to maturity (DTM) - Number of days from sowing to the point at which panicles of the plant turn brown in colour;
- Tiller number (TN) - Number of tillers on the date of heading;
- Culm length (CL) - Length (cm) from the base of the plant to the neck node of the main panicle;
- Panicle number (PN) - Number of panicles at maturity in each plant;
- Panicle length (PL) - Average length (cm) of the panicles in each plant;
- Spikelet number (SN) - Average number of spikelets, in each plant;
- Proportion of filled spikelets (PFS);

- Grain weight (GW) - Weight of 100 seed (g) at 14% moisture;
- Grain yield (GY) - Total weight (g) of the grain produced by a plant at 14% moisture;
- Dry matter (DM) - Weight (g) of the above ground parts (straw and grains) after oven drying at 70°C for 72 hours;
- Harvest index (HI) - The ratio between grain yield at 0% moisture level to the total dry matter of the plant.

RESULTS AND DISCUSSION

Mean of the basic and F₃ generation

The parents differed significantly for the majority of characters scored in both crosses except for proportion of filled spikelets (PFS), and grain yield (GY) in Cross 1 and for hundred grain weight (GW) in Cross 2 (Table 1). Tropical *japonica* parent was chosen because of low number of tillers, larger panicles and higher number of spikelets per panicle. As expected, Gaok, the tropical *japonica* variety was the lower scoring parent in the first pair of characters and the highest scoring parent for the latter pair in Cross 1. The Kemandy Pance, tropical *japonica* variety of the second cross was also chosen for the same but it did not show expected performances of lower number of tillers and panicles though it has larger panicles with more number of spikelets per panicle. Some of these differences between the parents, such as those for spikelet number (SN) in both crosses and, in particular for GY and dry matter (DM) and harvest index (HI) in Cross 2, are much larger than those commonly found in *indica* crosses (Perera *et al.*, 1997), which may be due to the fact that parents are from very different provenance.

F₁ means of the characters PN, GY and DM in Cross 1 are significantly higher than the better parent *i.e.*, *japonica* parent, indicating that heterosis is present for PN, GY and DM characters of Cross 1. There is very little evidence that heterosis is present in Cross 2. As the type of heterosis present indicates that the genes controlling the characters are dispersed between their parents of the cross, it is a good indicator for breeders to judge the potential of the cross in advance.

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Table 1. Mean performance of the families of the basic and the F₃ generations of each cross.

Character	P ₁	P ₂	F ₁	F ₂	F ₃	BC ₁	BC ₂
Cross 1							
DH	78.1 ²	63.8	69.6	70.3	70.5	74.3	67.0
DTM	110.4 ²	94.7	103.1	105.1	103.5	109.3	101.6
TN	8.6 ¹	4.9	9.8	8.6	7.3	8.6	8.5
CL	115.7 ²	72.2	95.6	95.9	91.9	108.1	84.6
PN	9.0 ¹	5.2	10.9	9.2	8.1	9.4	9.8
PL	31.4 ²	20.0	26.1	25.6	25.2	28.1	22.8
SN	211.4 ²	120.0	171.4	169.5	151.6	184.0	154.5
PFS	84.9 ¹	80.9	89.9	85.4	85.4	87.8	81.9
GW	2.9 ²	2.1	2.6	2.6	2.6	2.8	2.4
GY	22.6 ²	19.5	41.7	30.2	25.2	35.8	29.8
DM	47.6 ²	30.9	66.2	54.6	44.3	66.3	47.1
HI	56.3 ¹	41.8	54.5	48.3	51.2	54.9	47.3
Cross 2							
DH	95.2 ⁴	65.4	71.7	65.4	77.0	80.9	65.4
DTM	132.9 ⁴	94.3	97.2	104.8	113.0	117.2	97.9
TN	10.6 ⁴	5.3	7.9	8.3	8.9	10.2	7.1
CL	101.4 ⁴	81.6	94.0	100.9	98.9	112.3	96.3
PN	12.4 ⁴	5.3	7.9	8.4	9.3	12.2	7.4
PL	26.7 ⁴	18.7	21.8	23.0	24.4	27.3	21.0
SN	173.0 ⁴	116.5	146.3	116.3	170.5	179.2	124.2
PFS	82.8 ³	71.9	85.1	59.7	72.8	71.2	42.4
GW	2.41 ³	2.4	2.4	2.4	2.4	2.5	2.6
GY	35.1 ⁴	11.6	24.2	16.3	26.0	22.4	15.8
DM	100.8 ⁴	19.5	40.7	38.8	56.9	78.0	32.3
HI	55.3 ³	29.9	53.2	38.0	43.6	44.1	24.5

Superscript of the P indicate the parental variety involved where 1 = Jinmibeo, 2 = Gaok, 3 = Sangnambatbyeo and 4 = Kemandi Pance.

When proportion of filled spikelets (PFS) in generations was considered, there is a drastic drop in filled grains particularly in F_2 and B_2 in Cross 2. Usually *indica* cultivars consist of spikelets that are 80% filled and the objective of the NPT programme is to produce plants consisting of spikelets that have 100% filled grains. This criteria will have to be considered seriously when advancing crosses further in the breeding programme.

Though we expect the F_2 mean to be greater than F_3 mean, according to the usual genetical expectations (Mather and Jinks, 1982) in the absence of negative heterosis, which is marginally (though not significantly) the case with grain weight (GW) in Cross 2, inspection of Table 1 reveals that days to heading (DH), GW, and PFS in Cross 1 and most of the characters except culm length (CL) and panicle number (PN) in Cross 2 had greater F_3 mean than F_2 mean. This kind of effect may be due to the higher seed quality of F_3 produced from unclipped spikelets when compared with the rest of the seeds.

The component of basic generation means

The component of means were separated by weighted least squares procedure (Mather and Jinks, 1982). The satisfactory models (Table 2) in which all components were significantly different from zero and the χ^2 testing, the goodness of fit between the observed and expected values of the generation means, which were not significant could be found for all characters in Cross 1. Simple additive model (m and $[d]$) or additive and dominance model (m , $[d]$ and $[h]$) was sufficient for all characters except for related characters tiller number (TN) and panicle number (PN) for which a $[j]$ component (additive \times dominance non-allelic digenic interaction) was also required. The mean component $[h]$ was greater than $[d]$ for TN, PN, proportion of filled spikelets (PFS), grain yield (GY) and dry matter (DM), suggesting that the genes controlling these characters were dispersed between parents. In the Cross 2, the inadequacy of a simple m and $[d]$ or m , $[d]$ and $[h]$ model for most of the characters, except for TN and days to maturity (DTM), indicates the presence of epistasis which, for six of them, appears to be of the duplicate type as indicated by the opposite signs of $[h]$ and $[l]$ (Table 2). Average panicle length (PL) required a six parameter model, hence, test of adequacy of the model was not possible. Furthermore, it was initially not possible to find a satisfactory model for either spikelet number (SN) or DM. The inspection of Table 1 reveals that there were marked differences between generations for PFS.

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Table 2. Components of mean by weighted least squares procedure.

Character	m	[d]	[h]	[l]	[j]	[i]	[e]
Cross 1							
DM	70.5	7.0					
DTM	102.8	8.0	4.0				
TN	6.82	1.86	3.29		-1.70		
CL	95.8	22.7					
PN	7.21	1.91	4.08		-1.99		
PL	25.6	5.6					
SN	168.6	40.8					
PFS	81.5	3.9	8.3				
GW	2.56	0.39					
GY	21.2	2.2	20.4				
DM	40.3	10.7	27.3				
HI	48.0	7.4	5.5				
Cross 2							
DM	50.1	15.1	39.6	30.2			-18.0
DTM	113.8	19.3	-6.0				
TN	8.34	2.97					
CL	91.5	9.9	44.7		5.7		-42.2
PN	12.06	3.75	-4.06	-3.03			
PL	18.2	4.0	15.6	4.5	2.2		-12.1
SN	132.9	19.1	129.9	115.7			-1.3
PFS	77.3	5.40	-86.3		22.8	94.1	
GW	1.65		2.15	0.74			-1.43
GY	8.4	10.6	16.5	14.4			
DM	55.5	34.7	35.4	44.4			-0.5
HI	42.6	12.7	-38.9		7.0	49.4	

To overcome this problem, the generation mean was regressed onto PFS and a model was found with an additional parameter, e , which can be regarded as a dummy variable for SN and DM. This had the desired effect with both SN and DM, though the χ^2 testing for the adequacy of the model with respect to the latter was still just significant. It is surprising that this has not been the case with the other related characters, such as GY and HI. The phenomenon that $[h] > [d]$ for days to heading (DH), CL, PN, panicle length (PL), SN, PFS, GW, GY, DM and HI suggest that the genes controlling these characters are dispersed between parents, which suggest that the recombinant inbred lines that can be extracted from these crosses should show transgressive variation.

Table 3. Analysis of variance of F_3 families.

Character	Cross 1		Cross 2	
	MSb	MSw	MSb	MSw
df	29	197-201	29	206-209
DH	51.0***	13.3	343.3***	79.0
DTM	106.1**	50.7	735.3***	134.3
TN	8.4*	5.0	28.3***	7.2
CL	469***	110	758***	123
PN	16.9***	8.6	38.5***	10.7
PL	29.9***	4.4	29.9***	5.6
SN	4250***	759	6123***	1490
PFS	240***	136	1000***	204
GW	0.43***	0.15	0.14***	0.05
GY	154ns	109	359***	133
DM	606**	328	3145***	645
HI	183ns	137	846***	149

ns denotes non significance
* denotes significance at 1%

** denotes significance at 0.1%
*** denotes significance at 0.01%

Analysis of F₃ families

One way analysis of variance using data from F₃ families for both crosses shows that there are significant differences between families for all the characters except grain yield (GY) and harvest index in (HI) Cross 1 (Table 3).

It is clear, therefore, that apart from these two, all other characters are heritable. The failure to detect heritable variation for HI for this cross, even though the parents differed significantly for the character, could be due to the fact that these values are derived as the ratio of two primary characters ($HI = GY/DM \times 100$), which could be subjected to a high sampling variance. The estimates of heritabilities of these characters varied from low to high (Table 4).

Table 4. Narrow sense heritabilities.

Character	Cross 1	Cross 2
DH	0.40	0.47
DTM	0.11	0.60
TN	0.08	0.27
CL	0.26	0.40
PN	0.18	0.20
PL	0.65	0.46
SN	0.40	0.50
PFS	0.07	0.15
GW	0.36	0.14
GY	0.05	0.11
DM	0.12	0.77
HI	0.04	0.39

Potential of the crosses

The proportions of superior recombinant inbred lines that can be extracted with the desired NPT target or the desired interval for each cross was predicted according to Jinks and Pooni (1976) and is tabulated in Table 5. The experiment was conducted in 30 × 20 cm spacing, under transplanted condition, a target of 8-10 productive tillers (for TN and PN) has been used as a reasonable compromise for the original target of 3-4 tillers. At this density, for a crop to achieve a yield of 13 t/ha, each plant would have to produce 78 g of grain. The standards for PL, GW and DM were not specified for NPT; these have been taken, therefore, as equivalent to the score of the best parent (>P₁).

Table 5. Percentage of predicted recombinant inbred lines.

Character	Target	m	D	P%	m	D	P%
		Cross 1			Cross 2		
DH	70-80	70.5	9.96	56	77	66.4	45
DTM	100-130	103.5	14.65	82	113	151.0	77
TN	8-10	7.26	0.90	22	8.85	5.32	34
CL	90-100	91.9	95.12	37	98.9	159.8	30
PN	8-10	8.10	2.26	42	9.27	7.11	29
PL	≥ 31.4	25.5	6.88	1	24.4	6.21	17
SN	200-250	151.6	942	6	170.5	1182	9
PFS	1	85.4	28	0	72.8	203.1	3
GW	≥ 2.89	2.56	0.076	18	2.39	0.024	45
GY	≥ 78	25.2	13.4	0	26	57.1	0
DM	≥ 47.6	44.3	75.0	49	56.9	633.8	4
HI	≥ 0.6	51.2	-	-	43.6	176.7	11

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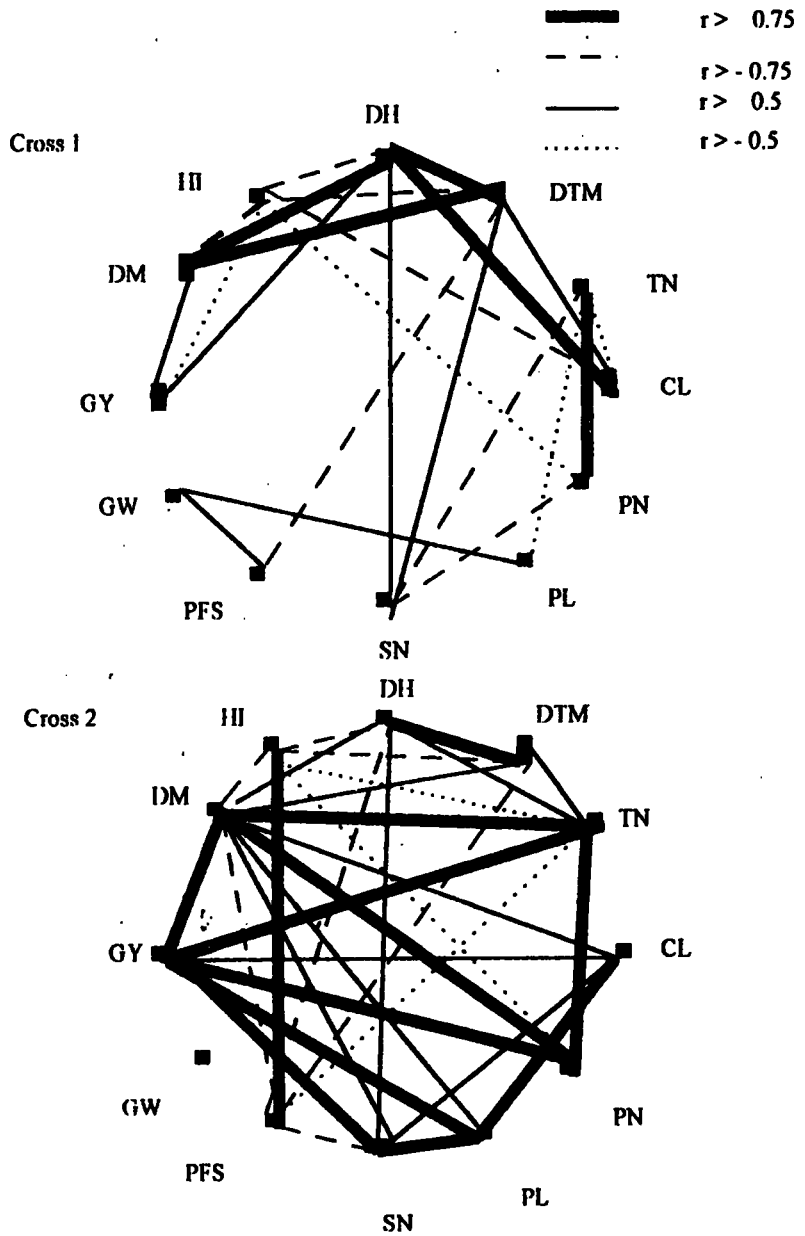


Figure 1. High and significant genetic correlations (r) between characters from F_2 family means (Mode and Robinson, 1959).

The evidence presented by Perera *et al.* (1992) and Fahim (1995) in rice suggested that the actual performances agreed best with the F_3 predictions which are more relevant to plant breeders. Therefore the predictions (Table 5) have been made for each character in each cross by using estimates of the mid parent (m) and additive genetic variance (D) from F_3 families. These results indicate that it could be fairly easy to extract recombinant inbred lines from this cross for which the performance achieved the NPT targets for days to heading (DH), days to maturity (DTM), tiller number (TN), culm length (CL), panicle number (PN) and spikelet number (SN) but that it would be more difficult to adopt the same for proportion of filled spikelets (PFS) and nearly impossible, with one cycle of inbreeding, to achieve the targets for grain yield (GY) and harvest index (HI).

Several hundred lines would have to be raised in order to find crosses for which the performance exceed the better parent for panicle length (PL). Since there is no evidence that HI is heritable in Cross 1, it appears impossible to achieve the desired target for this character. The results in Cross 2 indicate that it should be relatively easy to extract recombinant inbred lines from this cross whose performance achieve NPT targets for DH, DTM, TN, CL, and PN but is difficult with PFS and nearly impossible to achieve the NPT target for GY with one cycle of inbreeding. Therefore, neither of these crosses appear to offer much promise with regards to the yield improvement and proportion of filled spikelets. Apart from special cases such as breeding for some pest and disease resistance, plant breeders are, often concerned with improving their crops for several characters simultaneously. A knowledge of the genetical correlation between characters can, however, indicate, in a quantitative sense, at least, how likely a breeder is to meet the desired target for several characters at a time. The absolute values of genetic correlations greater than 0.5 of which the values which are significantly different from zero are presented in Figure 1 for both the crosses. Some of these correlations such as those between DH, DTM, TN and PN, for which $r > 0.75$ in both crosses involve characters which have an obvious pleiotropic relationship. Since, however, the pattern of correlation in Cross 1 is very different from that of Cross 2, the chief cause of genetic correlation in these crosses appear to be due to the linkage disequilibrium of genes that are linked in their inheritance.

CONCLUSIONS

The genetical architecture of the NPT characters across the two crosses were dissimilar. The characters of Cross 1 displayed mainly additive or additive and dominance effects while the characters of Cross 2 display epistasis of duplicate kind. There was significant genetical variation existing in all characters in both crosses except GY and HI in Cross 1. Heritabilities of these characters varied from low to high. From the investigation of the potential of crosses, it is not possible to achieve the NPT target for GY and PFS in either cross with only one cycle of inbreeding, though it is easy to meet the NPT target for DH, DTM, TN, CL and PN in both crosses. The genetic correlation in these crosses appear to be due to the linkage disequilibrium of genes that are linked in their inheritance. The most striking feature of the results obtained from the second cross was that there was a marked loss of fertility in terms of proportion of filled spikelets, in the F₂ and back cross generations, which is also a major problem with inbred lines developed by NPT programme.

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REFERENCES

- Dingkuhn, M., Penning de Vries, F.W.T., De Datta, S.K. and Van Laar, H.H. (1991). Concepts for a new plant type for direct seeded flooded Rice in the Tropics. *In: Direct seeded Flooded Rice in the Tropics*. IRRI, Los Banos, Philippines. pp. 17-38.
- Fahim, M. (1995). Quantitative inheritance of agronomic characters and comparison of different breeding methods in rice. Ph.D. thesis, University of Birmingham, U.K. pp. 89.
- Glaszmann, J.C. (1987). Isozymes and classification of Asian rice varieties. *Theor. Appl. Genet.* 74: 21-30.
- Herera, R.M. and Coffman, W.R. (1974). Emasculation of rice by vacuum extraction. *Proc. Crop Sci. Soc., Philippines*. 5: 12-14.
- IRRI. (1989). Work plan for 1990-1994. IRRI, Los Banos, Philippines. pp. 16.
- Janoria, M.P. (1989). A basic plant ideotype for rice. *Int. Rice Res. Newsletter*. 14: 12-13.

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- Jinks, J.L. and Pooni, H.S. (1976). Predicting the properties of recombinant inbred lines derived by single seed descent. *Heredity*. 36: 253-266.
- Mather, K. and Jinks, J.L. (1982). *Biometrical Genetics*. Chapman and Hall, London (3rd Edition).
- Mode, C.J. and Robinson, H.F. (1959). Pleiotropism and the genetic variance and covariance. *Biometrics*. 15: 581-537.
- Perera, A.L.T., Fahim, M., Sriyoheswaran, S., Dhanapala, M.P., Senadhira, D. and Lawrence, M.J. (1997). Quantitative genetics of rice. 1. No evidence of a shortage of exploitable genetical variation in modern *indica* cultivars. *Field Crop Res.* (submitted).
- Perera, A.L.T., Palihawadana, S. and Lawrence, M.J. (1992). Testing the predicted performance of recombinant inbred lines in rice using Triple Test Cross (ITC) design, Basic generations and F₃ families. *Int. Rice Res. Newsletter*. 17(6): 5.
- Vergara, B.S. (1988). Raising the yield potential of rice. *Philipp. Tech. J.* 13: 3-9.