INHERITANCE OF SPIKELET FERTILTY IN TWO RICE CROSSES

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ABSTRACT

Spikelet sterility is the greatest barrier to rice hybridization. Two crosses IDSA 85/NERICA 1 and IDSA 85/JASMINE 85 were made to study the inheritance of spikelet fertility. Percent Spikelet fertility was obtained by calculating the number of filled spikelets as a percentage of the total number of spikelets. F_2 spikelet fertility was then ranked into "low fertility class" plants and "normal fertility class" plants based on the fertility range of the less fertile parent. Any plant that fell into a fertility class similar to the range of fertility classes that the less fertile parent fell into was regarded as having a normal fertility and those that did not fall into this range were regarded as having a low fertility. There were significant differences ($p<0.001$) in mean fertility amongst the various populations in both crosses. Parents had significantly higher mean percent fertilities than F_1 and F_2 populations for both crosses. Mean percent fertility in F_2 was significantly higher than F_1 for the IDSA 85/NERICA 1 cross but there was no difference in F_1 and F_2 for the IDSA 85/ JASMINE 85 cross. The F_2 of the two crosses made in this study were found to have segregated into 9 low fertility plants: 7 normal fertility plants. This indicates that spikelet fertility in both crosses displayed duplicate recessive epistasis.

Keywords: *Rice, Inheritance, Spikelet fertility/ sterility, Hybridization, Duplicate recessive epistasis*

INTRODUCTION

Rice is now an important staple in Ghana with a per capita consumption of 14.5 kg per annum and consumption increasing at a rate of 20% per annum (Conway, 1999; MOFA, 2003). Although Ghana has the potential to be self sufficient in rice production, the current production level is only 35% of its consumption. About 380,000 metric tonnes of rice is imported into the country every year to offset the deficit (GPHA, 2004; Conway, 1999). This is because the local farmers are unable to cope with the stiff competition posed by rice imported into the country.

Consumers have consistently complained about the low quality of locally produced rice. Apart from the presence of foreign matter and lack of good mills, which result in badly polished rice,

most of the problems associated with locally produced rice are genetic and can be manipulated by breeders. For example, some of the qualities that consumers look out for include presence of aroma, extra-long/long and slender grains and intermediate amylose content, which results in rice which cook moist and tender, and do not become hard on cooling (Khush *et. al*, 1979). It is unusual to get all the qualities that consumers look for in a single high yielding variety adapted to the prevalent biotic and abiotic stresses. It is, therefore, important to combine the good traits from different cultivars/ lines into a single variety. The resultant variety must be high yielding, adaptable to the environment, disease resistant and possess acceptable cooking and eating qualities. This can be achieved through hybridization.

The greatest barrier to rice hybridization is F_1 hybrid sterility. Hybrid sterility results from disharmonious interactions between nuclear genes or between cytoplasm and nucleus in addition to differences in the structure of chromosomes. This phenomenon is more pronounced in interspecific hybridization than in intra-specific hybridization. Infertility limits procedures for obtaining gene recombination and limits seed production. For example, interspecific crossing of the African indigenous rice *Oryza glaberrima* with *Oryza sativa* cultivars is hindered by crossing barriers causing 100% spikelet sterility in F_1 hybrids (Heuer and Miezan, 2003). In general, hybrid fertility of intra-subspecific crosses, i.e., indica by indica (I x I) and japonica by japonica (J x J), is much higher than inter-subspecific crosses (I x J or J x I) (Liu *et al.*, 1996).

Spikelet sterility in F_1's may also be due to pollen sterility or anther indehiscence (Maekawa *et al.*, 1997; Sano, 1997). Various techniques like anther culture and embryo rescue have been used to overcome fertility barriers to rice hybridization (WARDA, 2000).

Information on the inheritance of spikelet sterility/fertility will facilitate the development of breeding strategies to complement the biotechnological methods that are being used to overcome fertility problems associated with rice hybridization. The objective of this work was therefore to determine the inheritance of spikelet fertility/sterility in crosses involving IDSA 85/ NERICA 1 and IDSA 85/JASMINE 85.

MATERIALS AND METHODS

The study was conducted at the experimental sites of the Crops Research Institute at Kwadaso and Ejisu-Besease (6°45'N, 1°25'W) from October 2001 to December, 2003. The experiment at Kwadaso was done in pots, whilst that at Ejisu-Besease was done in the field. Rice cultivars used for the experiment were IDSA 85, developed in the Côte d'Ivoire from Araguaia/ Cuiabana cross and released in Guinea in 1998; WAB 450-I-B-P-38-HB also known as Nerica 1 (N1), developed from interspecific hybridization between *Oryza sativa* (WAB56-104) and *O. glaberrima* (CGI4) and Jasmine 85(J85), which was developed by IRRI from IR 262/Khao Dwak Mali 105, and released in the USA in 1989.

Healthy and uniform seeds of the three varieties were sown; sowing was staggered over a two-week period in order to synchronize flowering in the three varieties. Thirty seeds each of the three varieties, were first pre-germinated on "Rose" paper tissue for four days, and then sown in plastic pots filled with sandy loam, which served as nursery pots. Twenty plants each of the three varieties were transplanted into individual pots (one plant/pot) after 21 days. The crosses made were IDSA 85/N1 and IDSA 85/J85. The F_2 seeds were obtained from the above crosses by allowing the F_1 ones to self. The IDSA 85/N1 cross was a pot experiment; pots were arranged adjacent to one another with a spacing of about 30cm x 30cm in between plants. IDSA 85/J85 was done in the field; seedlings were transplanted to the rain-fed lowland field at a single plant per hill spaced at 40 cm x 40 cm. Parent, F_1 and F_2 generations for each experiment were

planted at the same time to minimize environmental effects. Plants were taken care of under standard agronomic practices. For the pot experiment, 8 g of N.P.K (15-15-15) fertilizer was applied to each pot at transplanting, during tillering and at panicle initiation. For the field experiment, the recommended fertilizer rate of 90, 60, 45 kg ha^{-1} N, P_2O_5 and K_2O was applied. Foliar fertilizer, foliar calamax containing trace elements, was also applied on leaves at tillering. Weeds were controlled by hoeing and hand picking. Plants were watered whenever necessary and scaring of birds was done to eliminate bird damage.

Fertile spikelets were identified by pressing the spikelets with the fingers and noting those that were filled. The fertile and sterile spikelets were counted manually. Number of plants measured were 15 parents and 15 F_1 for both crosses; 197 and 855 F_2 for IDSA 85/N1 and IDSA 85/J85 crosses respectively. Percent Spikelet fertility was obtained by calculating the number of filled spikelets as a percentage of the total number of spikelets. Percent spikelet fertility was ranked according to the ranking of INGER (1996) as follows; highly fertile (≥ 90%), fertile (75-89%), partly sterile (50-74%), highly sterile (< 50% to trace) and completely sterile (0%). F_2 spikelet fertility was then re-ranked using the method of Maekawa *et al.* (1997). Any plant that fell into a fertility class similar to the range of fertility classes that the less fertile parent fell into was regarded as having a normal fertility and those that did not fall into this range were regarded as having a low fertility. Data was transformed using Arc sine and subjected to analysis of variance (ANOVA) with GENSTAT 7.1 statistical package. Observed F_2 segregation ratios for low fertility/normal fertility phenotypes were compared to various genetic ratios and the agreement or otherwise of these ratios were tested by means of the Chi-square test.

RESULTS

IDSA 85 was found to be more fertile than N1 and J 85 which had similar fertility levels (Tables 1 and 2). For the IDSA 85/N1 cross (Table 1), spikelet fertility was either fertile (75-89%) or highly fertile (≥ 90%) for the two parents except for N1 which had some plants being partly sterile (50-74%). All F_1, as well as 105 F_2 plants fell into the class, <50% to trace (highly sterile). Based on spikelet fertility of N1, F_2 plants that fell within partly sterile to highly fertile classes were regarded as having a normal fertility whilst those that fell below 50% were regarded as having low fertility (Maekawa *et al.*, 1997). Therefore, the F_2 population segregated into 105 plants with normal fertility and 92 plants with low fertility (Table 3). There was significant difference between mean percent spikelet fertility ($p < 0.001$) amongst the various populations. There was no difference between mean percent fertility in the two parents. However both parents had significantly higher mean percent fertilities than F_1 and F_2 populations. Mean percent fertility in F_2 (55.32%) was significantly higher than F_1 (25.92%) (Table 1).

For the IDSA 85/J85 cross (Table 2), IDSA 85 (parent) plants were either fertile or highly fertile. Spikelet fertility for J85 ranged from partly sterile to highly fertile. All the populations except F_2 had no plant which was completely sterile. All the 15 F_1 plants fell within the class <50% to trace. Based on the Spikelet fertility of J85, partly sterile to highly sterile plants in the F_2 were regarded as belonging to a "normal fertility group" whilst plants falling within the range highly sterile to completely sterile were regarded as belonging to a "low fertility group". Based on this criterion, F_2 segregated into 472 plants in low fertility group and 383 plants in normal fertility group (Table 3). There were significant differences ($p < 0.001$) in mean percent spikelet fertility amongst the different populations. Fertility in IDSA 85(92.80%) was significantly higher than J85 (83.56%). However, both parents had significantly higher fertilities than the F_1 (35.75%) and F_2 (45.59%) progenies. Mean fertility in F_1 and F_2 was not different for this cross (Table 2).

Table 1: Percent and Mean Percent Spikelet Fertility in Parents, F_1 and F_2 Populations in the IDSA 85/N1 cross

Fertility class (%)	Number of plants				
Population	**IDSA 85 (P_1)**	**N1 (P_2)**	**F_1**	**F_2**	**S.E.D**
0	0	0	0	0	
< 50 to trace	0	0	15	105	
50-74	0	2	0	25	
75-89	1	3	0	39	
≥ 90	14	10	0	28	
Total	15	15	15	197	
Mean Percent Fertility	1.33(93.74)	1.18(83.64)	0.53(25.92)	0.84(55.32)	0.85

0% = Completely Sterile
< 50% to trace = Highly Sterile
50-74% = Partly Sterile
75-89% = Fertile
≥ 90% = Highly Fertile

Table 2: Percent and Mean Percent Spikelet Fertility in Parents, F_1 and F_2 Populations in the IDSA 85/J85 cross

Fertility class (%)	Number of plants				
Population	**IDSA 85 (P_1)**	**J85 (P_3)**	**F_1**	**F_2**	**S.E.D**
0	0	0	0	13	
<50 to trace	0	0	15	459	
50-74	0	3	0	295	
75-89	3	10	0	85	
≥ 90	12	2	0	3	
Total	15	15	15	855	
Mean Percent Fertility	1.31(92.80)	1.16(83.56)	0.64(35.75)	0.73(45.59)	0.67

0% = Completely Sterile
< 50% to trace = Highly Sterile
50-74% = Partly Sterile
75-89% = Fertile
≥ 90% = Highly Fertile

Table 3: Summary of Genetic Analysis of Spikelet Fertility in the F_2 Populations of the Two Crosses

Types of Cross	Number of F_2 Plants		Ratio	χ^2	P values
	Low Fertility	Normal Fertility			
IDSA 85/N1	105	92	9:7	0.58	0.45
IDSA 85/J 85	472	383	9:7	0.34	0.56

A summary of genetic analysis of spikelet fertility is presented in Table 3. Both crosses segregated into 9 low fertility plants: 7 normal fertility plants.

DISCUSSION

Mean percent fertility as well as the various fertility classes in the F_1 and F_2 indicated that seed set in the F_1 was very low but at F_2 there was some restoration of fertility. Higher F_2 mean values than F_1 mean values may suggest the presence of epistatic and dominant genes (Kaw, 1999). Plants were re-grouped into a low fertility group and a normal fertility group based on the fertility range of the less fertile parent. Based on this re-grouping, the F_2 of both crosses were found to have segregated into 9 low fertility group plants: 7 normal fertility group plants; χ^2 = 0.58, P= 0.45 and χ^2 = 0.34, P= 0.56 for IDSA 85/N1 and IDSA 85/J 85 respectively (Table 3). The 9:7 ratios indicate that spikelet fertility in both crosses was under the control of two genes, showing duplicate recessive epistasis. This is in agreement with the findings of Virmani *et al.*1986; Zhu *et al.*(1998) and Wang *et al.* (2002). Virmani *et al.* (1986) reported that fertility was under the control of two genes and indicated F_2 segregation ratios of 9:3:4, 9:6:1 or 12:3:1 depending on the cross combinations. This result indicated that the interaction of the two genes was recessive epistasis, semiepistasis or dominant epistasis respectively. Zhu *et al.* (1998) reported that sterility caused by the cross of Akihikara and IR24 was controlled by two S-loci, S-5 and S-p(t), on chromosome 11. Two Quantitative trait loci (QTLs) for F_2 spikelet sterility were also detected on Chromosome 1 and 8 by Wang *et al.* (2002).

The 9:7 ratios also confirmed the involvement of epistatic genes in the inheritance of this trait (Jones and Karp, 1996).

It is recommended that adequate backcross populations be used to confirm the segregation ratios obtained. It is also recommended that anther culture techniques as well as wide compatibility varieties (WCVs) that produce normal fertility hybrids when crossed with wide range of varieties (Liu *et al*, 1996) be introduced into rice hybridization programmes in Ghana.

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