GENETICS OF MALE STERILITY IN RICE

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THE purpose of the work reported in this paper, was to determine the cause and nature of sterility of a highly sterile line of rice. It was characterized with regard to flower morphology, fertility, cytology and inheritance.

MATERIALS AND METHODS

A line of rice, G2266 from the intervarietal cross Bluebonnet 50 × Gulfrose, followed by selfing for six generations was identified as having reduced seed set. The standard variety Nato was used as fertile control plant, for comparison and for crossing with G2266. Nato (C.I. 8998), is a selection from the cross Rexoro-purpleleaf × Magnolia.

G2266 and fertile control plants were grown in the greenhouse, at the Agricultural Research and Extension Centre at Beaumont, Texas. Crosses between sterile and fertile control plants and subsequent back crossing utilized the standard clipping technique. Controlled selfing of parental plants was done by clipping the florets and bagging without removal of anthers.

The young tillers from each mother plant (parental and F_1) were separated and transplanted in field, thus vegetatively increasing the populations. Seeds from each F_1 plant and parental plant were sown in plots adjacent to the plot containing the transplanted tillers from F_1 plants and parental plants. Standard cultural practices were followed. Growing parental, F_1 and F_2 plants in one area in the field approached uniformity for comparison of floret fertility. Backcross progeny were grown in the green house. Fully ripened grains were harvested in panicles from the parent plants, the F_1 hybrids and the F_2 plants. Three panicles were harvested per plant and kept separate in bags.

Pollen fertility was estimated for experimental plants grown under field conditions. Meiosis was studied in pollen mother cells (PMC's) from temporary squash preparations using the acetocarmine technique.

The full and empty florets of the three panicles per plant were counted and recorded. As the segregating F_2 and backcross progeny showed different degree of sterility, it was necessary to classify them as follows: Sterile plants—showing 0–25% florets fertile; Semisterile plants—showing 26–75% florets fertile; Fertile plants—showing 76–100% florets fertile.

RESULTS AND DISCUSSION

In Nato, opening and closing of lemma and palea during blooming of the florets were regular. There were consistently six anthers in a floret and anthers were yellow at maturity. Dehiscence of anthers took place normally at blooming. The pistil consisted of a well developed ovary with a short style terminated by two plumose stigmas. Examination of pollen grains using acetocarmine technique revealed that more than 97% of them were stainable. Frequency of seed set was more than 86%.

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Blooming of florets in line G2266 was normal. Colour of anthers was white to pale yellow. Anthers, six in number per floret, were generally indehiscent, but with occasional dehiscence. The pistil was apparently normal. The frequency of acetocarmine-stainable pollen in G2266 was greatly reduced compared to the control fertile plant (Table 3). The frequencies of stainable pollen varied from 0% to 16% with a mean of 2% from anther to anther in a floret. Nonstaining pollen appeared small, transparent, colorless and empty grains. All the stages of meiosis in the PMC's appeared to be normal in G2266. Seed set in G2266 was much lower (2%) compared to control plants (87%).

When G2266 were pollinated by Nato, seed set was high (36%). When G2266 was used as the pollinator pollen parent, seed set was reduced to 11%. Seed set in G2266 was 8% and in Nato 25% on controlled selfing. These observations indicate that the pollen of G2266 was not as effective as pollen from Nato in producing seed on either G2266 or control plants. Since seed set, when G2266 was used as the female parent, was as high as or higher than for the selfed normal variety the ovules of G2266 appeared to be normal. Only the (Nato × G2266) F_1 seeds gave viable F_1 plants, but not the reciprocal F_1 seeds, which might be due to dormancy or perhaps fungal infection.

Floret sterility in F_1 plants and their parents.—Mean frequency of seed set was high in the fertile parent and low in the sterile parent (Table 1). However, the frequency of seed set in each hybrid approached the intermediate value between the mean frequency of the parents. The mean percent seed set in the hybrids was 50%. The fertile parent had a mean seed set frequency of 87%. The mean frequency of seed set in the sterile parent was 2%. Frequency of total florets per panicle in the F_1 hybrids was not considerably different from that in the fertile parent.

TABLE 1

Mean percentage seed set in F_1 plants (Nato × G2266) and their parents

F_1 Plant number*	Number of Plants**	Percent seed set	Phenotypic classification
1	1	48	semisterile
2	1	42	semisterile
3	1	54	semisterile
4	1	57	semisterile
5	2	52	semisterile
6	2	49	semisterile
Nato	10	87	fertile
G2266	7	2	sterile

*F_1–1 to F_1–6 are F_1's from Nato × G2266.
**Plants obtained by transplanting tillers from the F_1 and the parental plants.

It appeared that all F_1 plants from the cross Nato × G2266 were uniformly semisterile and thus were genotypically similar for floret sterility. Since all plants produced from selfed seeds of G2266 were highly sterile (mean seed set of 2%) it was likely that the parent G2266 was homozygous for this level of sterility.

Pollen development and characteristics in F_1 plants.—Study of pollen development in PMC's from the F_1 plants of the cross between G2266 and the fertile parent, using acetocarmine technique showed that meiosis was apparently normal. The frequency of stainable pollen varied from 13% to 98% between the F_1's (Table 2). Variation was present in the frequency of stainable

TABLE 2

Mean percentage stainable pollen in F_1 plants (Nato × D2266) and their parents

F_1 Plant Number	Flower number	Percent stainable pollen	Mean percent stainable pollen
1	1	99	
	2	54	
	3	40	64
2	1	97	
	2	99	
	3	100	98
3	1	42	
	2	64	
	3	98	68
4	1	40	
	2	0	
	3	0	13
5	1	96	
	2	8	
	3	10	38
6	1	100	
	2	98	
	3	71	90
Nato	1	99	
	2	98	
	3	99	98
G2266	1	3	
	2	3	
	3	2	2

pollen among flowers from a single F_1 plant. Variation was also found among anthers from the same flower. For example, the frequencies of stainable pollen in the 3 anthers in a flower from F_1-6 were 58%, 73% and 82%. Therefore, not all pollen in the F_1 plants was normal. The results however, did not indicate whether the apparently normal pollen in the hybrids was as fertile as those in the fertile control plants. Seed set did approach 50% in the F_1's so some normal pollen was produced.

F_2 seed set performance.—With a single exception (F_1-2) all F_1 hybrids showed segregation for sterility (Table 3). Results also showed that plants grown from selfed seeds of the fertile control parent were all fertile. Plants grown from the selfed seed from the sterile parent were all sterile. As selfed progeny from G2266 were all similar in sterility, it is probable that G2266 was homozygous for the factor controlling sterility and the F_1 hybrids were genotypically similar with respect to the factors controlling sterility.

TABLE 3

F_2 segregation for sterility in (Nato × G2266)

F_1 Plant	Number of F_2 plants**	Phenotypic classification
1	4	2 fertile, 2 sterile
2	1	1 fertile
3	10	4 fertile, 3 semisterile, 3 sterile
4	29	17 fertile, 10 semisterile 2 sterile
5	24	7 fertile, 6 semisterile, 11 sterile
6	21	14 fertile, 1 semisterile, 6 sterile
Nato	57	all fertile
G2266	31	all sterile

**F_2 plants obtained by seeds from the parental plants.

Since all the F_1 hybrids, with one exception, had similar responses the F_2 data were pooled. It was difficult from the F_2 data to determine which plants were homozygous for fertility and which were heterozygous. There was apparently an overlapping of classes. The steriles, however, were quite distinct. It seems that a single locus was controlling male sterility in G2266. The homozygous recessive plants were sterile. The homozygous dominant plants were fertile. The heterozygous plants were apparently intermediate in fertility but could not definitely be distinguished from the fertile plants. The F_1

and F_2 plants flowerd under slightly different environmental conditions. This may account for the increased frequency of seed set in F_2 heterozygotes as compared to the F_1 plants. The ratio was 65 fertile- semisteriles; 24 steriles (χ^2 0·24, P between 0·50 and 0·75). There is no evidence that cytoplasm was involved in the control of sterility in G2266.

Seed set in reciprocal backcrosses: Backcross results showed that the frequency of seed set was consistently higher when the control plants were used as the male parent (Table 4). As expected, seed set was lower when the F_1 plant was used as the pollinator, with two exceptions. No seeds were set when G2266 was used as the male parent.

TABLE 4

Percent seed set by reciprocal backcrosses between F_1 (Nato × G2266) and their parents

F_1 plant number	Parents			
	Nato female	Nato male	G2266 female	G2266 male
1	0% (10)	22% (37)	0% (6)	0% (12)
2	56% (9)	70% (10)	20% (34)	0% (15)
3	3% (31)	37% (35)	16% (6)	0% (12)
4	0% (7)	—	0% (12)	—
5	14% (22)	88% (8)	50% (16)	0% (10)
6	6% (29)	48% (37)	10% (19)	0% (9)

Number in parenthesis indicates number of flowers pollinated.

Results on seed set, in the reciprocal backcross between F_1 hybrids and their respective parents suggest pollen in G2266 was defective. Comparison of seed set in the backcrosses where fertile control parent and sterile G2266 were used as female parent and the F_1 plants as male parent showed that seed set was similarly affected (Table 4). These results indicated that ovules in G2266 were equally fertile as in the fertile control parents.

Results of seed set by reciprocal backcrosses, therefore confirmed results of seed set data in the original crosses between G2266 and the fertile control plant. This also confirmed the findings from the results on pollen examination carried out in the F_1 and the G2266.

SUMMARY

A line of rice (G2266) was found to be male sterile, When line G2266 was used as female parent in crosses with normal fertile plants seed development was normal. However, seed development was reduced (11% seed set) in the

reciprocal crosses. All F_1 hybrids were semisterile. Although meiosis in PMC's in G2266 and the F_1 hybrids was regular, the frequency of normally stained pollen grains was low in G2266 (2%). A wide range in stainable pollen (0–100%) was found in the F_1 hybrids. Backcrossing the F_1 hybrids as pollen parent caused reduction in seed development. The reciprocal backcrosses using the normal parent as pollinator gave rise to higher seed set. Seed set was also reduced in those reciprocal backcrosses which involved G2266 as the pollen parent. Segregation of approximately three fertile semisteriles to one sterile individual occurred in the F_2 generation, suggesting that male sterility was due to a recessive gene free of cytoplasmic influence.

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