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Genetics of fertility restoration in 'WA' cytoplasmic male sterility in rice (*Oryza sativa* L.)

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Abstract

A study using three cytoplasmic male sterile lines (IR-68897A, IR-68888A and IR-58025A) and ten restorers revealed that two or three major genes govern the fertility restoration, with epistatic interaction that differed from cross to cross. The crosses, IR-68897A x URG-22, IR-68888A x URG-51, IR-68888A x URG-84, IR-58025A x Sarjoo-52 and IR-58025A x URG-48 showed segregation ratio of 9:6:1 in F₂ and 1:2:1 in BC₁ generation, for pollen fertility indicating that the genotypes viz. Sarjoo-52, URG-22, URG-51, URG-48 and URG-84 carry two independent segregating genes with additive type of gene action, the effect of one of the two gene in restoring fertility appeared to be stronger than the other. The crosses IR-68888A x BPT-5204, IR-58025A x Pant-12 and IR-58025A x URG-77 segregated in ratio of 9:3:4 and 1:1:2 in F₂ and BC₁ population respectively, indicating the presence of two independently segregating dominant genes showing recessive epistatic interaction of the fertility restoring genes in the genotypes, BPT-5204, Pant-12 and URG-77. The crosses IR-68897A x Jaya and IR-68897A x Shivani given segregation behavior of 12:3:1 in F₂ progeny and 2:1:1 genetic ratio in test cross progeny suggesting the presence of two dominant genes with dominance epistasis in the fertility restoring genes in the genotypes Jaya and Shivani.

Keywords: Restorers, fertility restoration, hybrid

Introduction

Among the various approaches for improving the yield threshold of rice, exploitation of hybrid vigor is considered to be the most feasible and readily practicable. China pioneered hybrid rice research in 1970's and demonstrated 20-30% yield advantage over conventional varieties. The hybrid grown in India, China, Vietnam, Bangladesh and other countries are based on indica rice sources which on an average shows a standard heterosis of 15-20% in commercial cultivation. It has been demonstrated very clearly on large scale that hybrids give 15-20% increased yield over the highest yielding varieties under the similar growing conditions by using 'Wild Abortive' (WA) cytoplasmic male sterility.

High yield potential of CMS derived F₁ hybrids depends upon their high pollen and spikelet fertility which is determined by the number and mode of action of restorer genes present in the restorer parent. Knowledge of genetic control of male fertility restoration and extent of fertility restoration facilitates, transfer of fertility restorer genes to promising breeding lines and undertake improved restorer breeding programme and ultimately their deployment in hybrid breeding programme. The present investigation was undertaken to understand the genetics of fertility restoration of CMS lines of 'WA' cytoplasm by ten crosses.

Materials and Methods

Thirty rice genotypes were crossed with three 'WA' cytoplasmic male sterile lines viz. IR-68897A, IR-68888A and IR-58025A for the isolation of restorer lines during kharif-2010. Out of 30 only ten genotypes restored fertility in presence of cytoplasmic male sterile background, were identified by pollen and spikelet fertility at anthesis and maturity respectively during kharif-2011. At the same time the F₁'s were selfed and testcrossed (F₁ x CMS) for raising an appropriate plant population for genetic studies. During kharif-2012, F₁, BC₁F₁ and F₂ seeds of the restorers were raised having plant population of 150-200 for F₂ and 50-150 for BC₁F₁. Since spikelet fertility data did not give any convincing pattern as it is influenced by several physiological and environmental factors, the data on pollen fertility were considered reliable for the study. Pollen fertility studies were conducted using 0.5% iodine and 2% potassium

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iodide solution, where due care was taken about proper sampling from F₁, F₂ and BC₁F₁ populations. The pollen grains were squeezed out of the anther on a glass slide, treated with I₂KI solution and then observed under microscope. Fully, dark stained pollen grains were the fertile one where as unstained pollen grains were sterile. Plants were classified in to different fertility sterility groups as; more than 80 percent fertile pollens were grouped as fully fertile, 21-80 per cent as partial fertile, 1-20 per cent as partial sterile and less than one percent fertile pollen as completely sterile plants. The goodness of fit for various Mendelian genetic ratios in F₂ and test cross progenies (CMS x F₁s) was tested using the chi-square statistic.

Results and Discussion

In the present study, the inheritance of fertility restoration in the crosses IR-68897A x URG-22, IR-68888A x URG-51, IR-68888A x URG-84, IR-58025A x Sarjoo-52 and IR-58025A x URG-48, reveals F₂ segregation ratio of 9 (fertile): 6 (partial fertile + partial sterile): 1 (completely sterile), indicating that the genotypes *viz.* Sarjoo-52, URG-22, URG-51, URG-48 and URG-84 carry two independent segregating genes with additive type of gene action, the effect of one of the two gene in restoring fertility appeared to be stronger than the other because the presence of both the genes alone conferred partial pollen fertility/sterility (**Table-1**). The inference derived from the F₂ population was also confirmed from the segregation ratio the test cross progenies which exhibited segregation behavior 1(fertile): 2(partial fertile + partial sterile): 1(completely sterile). Such results are in agreement with the earlier findings of Li *et al.* (1993), Sohu and Phul (1995) [12], Ganeshan and Rangaswamy (1997) [2], Yang *et al.* (2002) [13] and Hossain *et al.* (2010) [4], who reported that two independently segregating dominant gene control fertility

restoration of 'WA' cytoplasmic male sterility in rice. F₂ population of the crosses IR-68888A x BPT-5204, IR-58025A x Pant-12 and IR-58025A x URG-77 segregated in ratio of 9(fertile): 3 (partial fertile + partial sterile): 4 (completely sterile) indicating the presence of two independently segregating dominant genes showing recessive epistatic interaction of the fertility restoring genes in the genotypes, BPT-5204, Pant-12 and URG-77. The test cross progenies exhibited the segregation ratio of 1 (fertile): 1 (partial fertile + partial sterile): 2 (completely sterile) plants, there by confirming the same. Thus, the plants homozygous for the recessive alleles of any of the two genes but homozygous or heterozygous for the dominant alleles of the other gene (R₁_r₂r₂ or R₂_r₁r₁) will be sterile depending upon which the two alleles are stronger or weaker. The crosses IR-68897A x Jaya and IR-68897A x Shivani given segregation behavior of 12 (fertile): 3 (partial fertile + partial sterile): 1 (completely sterile) in F₂ progeny and 2 (fertile): 1 (partial fertile + partial sterile): 1 (completely sterile) genetic ratio in test cross progeny indicating the presence of two dominant genes with dominance epistasis in the fertility restoring genes in the genotypes Jaya and Shivani. Similar observations suggesting recessive and dominant type interaction between two dominant fertility restoring genes were also reported by Govindraj and Virmani (1988), Bharaj *et al.* (1991) [1], Ramalingam *et al.* (1995) [7], Sarkar *et al.* (2002) [8], Sharma and Singh (2003) [9], Singh *et al.* (2008) [11] and Hossain *et al.* (2010) [4]. Kumari *et al.* (1998) [5] and Hossain *et al.* (2010) [4] reported that the differences in the type of gene interaction could presumably be due to variable expression of weaker gene in different genetic backgrounds. Penetrance and expressivity of the restorer gene in rice are known to be affected by the parental type of individual plants in segregating progenies (Singh, 1994) [10].

Table 1: Segregation pattern of pollen fertility restoration in F₂ and test cross population of the crosses of CMS lines with restorer lines

Cross combination	No. of plants with pollen fertility reactions in F ₂ population					Total plants scored	Genetic Ratio	Chi-square	p value	No. of plants with pollen fertility reactions in testcross population					Total plants scored	Genetic Ratio	Chi-square	p value
	FF	PF	PS	SF	CS					FF	PF	PS	SF	CS				
IR 68897A x Jaya	118	22	12	34	6	158	12:3:1	2.169	0.35	20	7	5	12	14	46	2:1:1	0.957	0.62
IR 68897A x Shivani	108	29	10	39	8	155	12:3:1	4.326	0.12	35	14	10	24	20	79	2:1:1	1.43	0.49
IR 68897A x URG-22	152	47	40	87	11	250	9:6:1	2.775	0.25	27	22	19	41	23	91	1:2:1	1.242	0.54
IR 68888A x BPT 5204	80	17	22	39	30	149	9:3:4	5.965	0.05	18	9	10	19	23	60	1:1:2	3.3	0.19
IR 68888A x URG-51	100	31	26	57	12	169	9:6:1	1.49	.32	27	14	22	36	27	90	1:2:1	3.6	0.17
IR68888A x URG-84	125	36	40	76	21	222	9:6:1	4.29	0.12	15	8	14	22	12	49	1:2:1	1.122	0.57
IR 68025A x Pant-12	92	18	17	35	36	163	9:3:4	1.199	0.55	18	12	10	22	52	92	1:1:2	1.913	0.38
IR 68025A x Sarjoo-52	88	36	30	66	12	166	9:6:1	0.79	0.67	17	12	18	30	13	60	1:2:1	0.533	0.77
IR 68025A x URG-48	68	19	25	44	11	123	9:6:1	1.546	0.46	17	20	16	36	20	73	1:2:1	0.26	0.88
IR 68025A x URG-77	118	22	19	41	46	205	9:3:4	0.771	0.68	8	3	2	5	22	35	1:1:2	2.829	0.24

FF=Fully fertile; PF=Partially fertile; PS=Partial Sterile; CS=Completely sterile; PF+PS= SF (Semi fertile)

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