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Full Length Research Paper

Genetics and combining ability of fertility restoration of 'wild abortive' cytoplasmic male sterility in rice

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Information on the genetics of fertility restoration facilitates breeding and/or selection of restorer lines used in hybrid breeding program in a cytoplasmic male sterility (CMS) system. Inheritance of fertility restoration of WA type CMS in rice, (*Oryza sativa* L.) was studied utilizing IR58025A, IR62829A and IR68899A CMS lines and three restorers viz., Amol-2, IR50 and Poya. The F1s showed pollen and spikelet fertility similar to the restorer parents, indicating that restoration ability were dominant and the cytoplasmic-genetic sterility system of CMS lines were sporophytic in nature. Evaluation of fertility in F2 populations and testcross progenies (BC1) revealed that fertility restoration in Amol-2 and IR50 were controlled by two major dominant genes whereas, it was controlled by one dominant gene in Poya. Segregation for spikelet fertility in F2 and backcross generation conformed to the results on pollen fertility. Analysis of line x tester indicated pre-dominance of non-additive genetic variance. It suggested greater importance of non-additive gene action in pollen and spikelet fertility expression and indicated very good prospect for the exploitation of non-additive genetic variation for pollen and spikelet fertility through hybrid breeding.

Key words: Rice, genetics, male fertility restoration, combining ability.

INTRODUCTION

Hybrid rice offers an opportunity to boost the yield potential of rice. It has a yield advantage of 15-20% over conventional high-yielding varieties (Virmani et al., 1993). Fertility restorer alleles (Rfs) are always tightly evolved with cytoplasmic male sterility (CMS) during plant evolution. Research of Rfs inheritance is the precondition for breeders to develop elite restorer lines. For studying the inheritance of the fertility restorers, in general, the main three indexes (percentage of fertile pollen, bagged seed setting and opening seed-setting) are often used as the criteria to evaluate fertility restoration. Of these, the percentage of fertile pollens is thought the most reliable criterion for evaluating plant fertility (Li et al., 2007).

Restorer lines are the critical factors for successful development of hybrid rice. With the scalable CMS-WA hybrid rice released for commercial production, the inheritance of Rf alleles have been extensively studied using various restorer lines with different origins. A number of reports about the inheritance of the restorer alleles for CMS-WA come from the restorer lines of IR24, IR64, IR8 (from IRRI), Minghui63, Milyang23 and Milyang46 that are widely used in commercial production. Different genetic models of Rf alleles such as one gene (Shen et al., 1996; Gyan et al., 2003), two linked genes (Li and Zhu, 1986; Guha Sarkar et al., 2002), and two independent genes (Li and Yuan, 1986; Virmani et al., 1986; Teng and Shen, 1994; Bharaj et al., 1995) are proposed by different groups. Yao et al. (1997) investigated the fertility segregation of plants in F2 of Zhen-shan97A/ Minghui63. They found that fertility segregation confirmed to 15:1 ratio, indicating that fertility restoration in this population is controlled by two major dominant genes. Gyan et al. (2003) found that the F2 of IR58025A x PRR-78, giving a good fit to monogenic 3:1 ratio. Fu and Xue (2004) analyzed the spikelet fertility of F1, F2 and BCF1 derived from five different crosses between the CMS-WA line and various restorer lines. They found that indica

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Abbreviations: CMS, Cytoplasmic male sterility; **Rfs,** restorer alleles; **GCA,** general combining ability; **F,** fertile; **PF,** partially fertile; **PS,** partially sterile; **CS,** completely sterile.

restorer lines Milyang46 and H804 possess two dominant restorer alleles, but japonica restorer lines of H921 and T984 have one restorer allele.

High yield of the F1 hybrids depends largely upon high pollen or spikelet fertility which is determined by the mode of gene action prevalent in the restorer lines of the hybrids. Knowledge of the genetic control of male fertility restoration is also useful to transfer fertility restoring genes to promising breeding lines and undertake improved restorer breeding programme. This study was undertaken to understand genetic control of fertility restoration in three promising varieties and breeding lines of rice in WA cytoplasmic genetic male sterility system using CMS lines IR58025A, IR62829A and IR68899A.

MATERIALS AND METHODS

Plant materials

Genotypes of rice used in this study were three CMS lines (IR58025A, IR62829A and IR68899A), and three restorer varieties accessions (Amol-2, IR50 and Poya). The three A lines all have cytoplasm derived from a WA cytoplasmic source. Crosses were carried out in all combinations between CMS lines and pollen parents. The F1 plants of these crosses were used as pollinators for the respective CMS lines to develop testcross progenies. Seedlings of the parental, F1, F2 and testcross progenies were planted 20 x 20 cm apart using single seedling per hill.

Estimation of pollen and spikelet fertility

Pollen fertility and seed-setting rate were used as the main criteria for the evaluation of fertile and sterile plants. Natural seed setting rates of F1 hybrids were used as the main criteria for the evaluation of compatibility. At least three different plants must be tested for every hybrid combination (Tan et al., 2008). For all materials, pollen grain fertility was measured using anthers collected from spikeletes at 1 to 2 days before anthesis and fixed in acetic acid-alcohol (1:3) solution (Sarial and Singh, 2000). Three undehised spikelets were randomly selected from different positions on the panicle. The anthers from each spikelet were smeared in a drop of 1% I2-KI solution on a glass slide separately and observed under compound microscope. Plants were classified into different fertility-sterility groups based on proportion of stained-round pollen grains, as the number of fertile grains/total number observed x 100. With regard to pollen fertility of testcross and F2 progeny, plants were classified into the following classes: Fertile (F): Plants showing more than 70% pollen fertility; partially fertile (PF): plants showing 50 to 70% pollen fertility, partially sterile (PS): plants showing 1 to 50% pollen fertility and completely sterile (CS): plants showing no pollen fertility (Gyan et al., 2003). The panicles that emerged from the primary tiller were bagged before anthesis and the number of filled grains and chaffs in the panicle were counted at the time of maturity. The ratio of filled grains to the total number of spikelets was expressed as seed setting rate (He et al., 2006). The goodness of fit to Mendelian segregation pattern of fertile, partially fertile, partially sterile and completely sterile plants in F2 and testcross populations was tested by Chi-square technique.

Line x tester analysis

The CMS lines IR58025A, IR62829A, and IR68899A were crossed with three elite varieties Amol-2, IR50 and Poya to generate 9 hybrid combinations in line x tester mating design. The hybrids were evaluated along with parents in a randomized complete block design with three replications in the Agricultural sciences and Natural Resources University, Sari, Iran, during 2007 - 2008. Thirty-day-old seedlings were planted (20 x 20 cm apart using single seedling per hill). Combining ability analysis was carried out following the method of Kemplhorne (1957).

RESULTS

Genetics of fertility restoration

Amol-2, IR50 and Poya varieties were identified as effective restorers of cytoplasmic genetic male sterile lines possessing WA cyto-sterility system (namely IR58025A, IR62829A and IR68899A). The inheritance of their fertility restoration ability was studied in the crosses. The data on pollen and spikelet fertility of the CMS lines, pollinators and F1 hybrids are presented in Table 1. Mean performance of lines, testers and their hybrids (Table 1) indicated worth of genetic variability for the study of gene effects on pollen and spikelet fertility, which are important in hybrid rice yield. The results revealed that the CMS lines IR58025A, IR62829A and IR68899A were completely sterile, whereas the pollinators Amol-2, IR50 and Poya were fertile. The pollen and spikelet fertility of the F1 hybrids ranged from 70.11 to 90.22% and 81.71 to 93.20%, respectively, thereby indicating that fertility restoration in these pollinators (Amol-2, IR50 and Poya) were under dominant gene control and the cytoplasmic-genetic sterility system of IR58025A, IR62829A and IR68899A were sporophytic in nature. The data on pollen fertility of F2 plants are given in Table 2. The results revealed that F2 populations of IR58025A/Amol-2, IR58025A/IR50, IR62829A/Amol-2, IR62829A/IR50, IR68899A/Amol-2 and IR68899A/IR50 crosses exhibited 9 fertile : 3 partially fertile : 3 partially sterile : 1 completely sterile plants, ($\chi^2 = 1.69 - 6.62$), indicating that restorer varieties Amol-2 and IR50 each carried two independently segregating dominant genes with additive effects for restoring fertility of the WA cytoplasm. The effect of one of the two genes in restoring fertility appeared to be stronger than the other. Assuming that R1 and R2 are the dominant alleles of the two restorer genes, the plants having dominant alleles of the two genes in homozygous or heterozygous condition (R1-R2-) will be fertile. The plants having dominant alleles of one of the two genes in homozygous or heterozygous condition but homozygous recessive alleles of the other gene (Rl-r2r2 or rlr1R2-) will behave partially sterile or partially fertile and vice-versa. The plants homozygous for the recessive alleles of both genes (r1rlr2r2) will be completely sterile. The inference derived from the F2 data on pollen fertility was confirmed from the segregation behavior of spikelet fertility (Table 3). Also, these results were confirmed from the segregation behavior of pollen and spikelet fertility of the testcross progenies

Table 1. Means of spikelet and pollen fertility of parents and F1 hybrids.

Parents or F1 crosses	Spikelet fertility (%)	Pollen fertility (%)
Lines		
IR58025 A	0.00	0.00
IR62829 A	0.00	0.00
IR68899 A	0.00	0.00
Testers		
Amol-2	93.20	89.46
IR50	86.10 *	73.67 *
Poya	91.10	85.59
Grand Mean	90.13	82.91
S.E.	1.08	2.40
C.D. (0.05)	3.32	8.01
C.D. (0.01)	4.36	10.5
Hybrids		
IR58025A/Amol-2	92.30 **	81.86
IR58025A/IR50	85.91 **	82.69
IR58025A/Poya	89.62	70.96 **
IR62829A/Amol-2	91.70 *	86.51
IR62829A/IR50	84.11 **	81.61
IR62829A/Poya	92.00 *	90.22 **
IR68899A/Amol-2	93.10 **	89.21 **
IR68899A/IR50	81.71 **	70.11 **
IR68899A/Poya	90.70	89.40 **
Grand Mean	89.02	82.51
S.E.	0.76	1.42
C.D. (0.05)	2.35	4.76
C.D. (0.01)	3.09	6.25

* and **; Significant at p = 0.05 and p = 0.01 levels, respectively, based on an F-test. C.D. = critical difference.

Table 2. Pollen fertility of plants in F2 populations.

Cross	Pollen fertility (%) Mean ± Se	Number of plants					Segregation ratio
		Total	F	PF	PS	CS	
IR58025A/Amol-2	72.61 ± 1.52	270	143	61	55	11	$\chi^2_{15:1} = 2.18^{ns}$; $\chi^2_{9:3:3:1} = 5.06^{ns}$
IR58025A/IR50	63.44 ± 1.56	269	138	64	50	17	$\chi^2_{15:1} = 0.002^{ns}$; $\chi^2_{9:3:3:1} = 4.82^{ns}$
IR58025A/Poya	44.03 ± 1.71	310	79	78	86	67	$\chi^2_{3:1} = 1.89^{ns}$; $\chi^2_{1:2:1} = 1.97^{ns}$
IR62829A/Amol-2	63.69 ± 1.30	271	164	48	45	14	$\chi^2_{15:1} = 0.54^{ns}$; $\chi^2_{9:3:3:1} = 2.21^{ns}$
IR62829A/IR50	65.71 ± 1.60	333	175	57	79	22	$\chi^2_{15:1} = 0.072^{ns}$; $\chi^2_{9:3:3:1} = 5.74^{ns}$
IR62829A/Poya	44.00 ± 1.80	285	61	77	83	64	$\chi^2_{3:1} = 0.98^{ns}$; $\chi^2_{1:2:1} = 4.36^{ns}$
IR68899A/Amol-2	66.98 ± 1.40	272	161	54	45	12	$\chi^2_{15:1} = 1.57^{ns}$; $\chi^2_{9:3:3:1} = 6.62^{ns}$
IR68899A/IR50	65.06 ± 1.38	328	176	69	66	17	$\chi^2_{15:1} = 0.64^{ns}$; $\chi^2_{9:3:3:1} = 1.69^{ns}$
IR68899A/Poya	46.18 ± 1.50	568	121	162	126	159	$\chi^2_{3:1} = 2.71^{ns}$; $\chi^2_{1:2:1} = 5.19^{ns}$

F (fertile) = > 70% Pollen fertility; PF (partially fertile) = 50 - 70%; PS (partially sterile) = 1 - 50%; CS (completely sterile) = 0.

Table 3. Spikelet fertility of plants in F2 populations.

Cross	Spikelet fertility (%) Mean ± Se	Number of plants					Segregation ratio
		Total	F	PF	PS	CS	
IR58025A/Amol-2	72.85 ± 1.90	270	154	59	44	13	$\chi^2_{15:1}=0.95^{ns}$, $\chi^2_{9:3:3:1}=3.17^{ns}$
IR58025A/IR50	62.34 ± 1.60	269	133	59	56	21	$\chi^2_{15:1}=1.11^{ns}$, $\chi^2_{9:3:3:1}=5.32^{ns}$
IR58025A/Poya	47.68 ± 2.01	310	71	87	83	69	$\chi^2_{3:1}=1.24^{ns}$, $\chi^2_{1:2:1}=2.8^{ns}$
IR62829A/Amol-2	77.93 ± 1.61	271	158	55	42	16	$\chi^2_{15:1}=0.05^{ns}$, $\chi^2_{9:3:3:1}=2.13^{ns}$
IR62829A/IR50	67.70 ± 2.00	333	186	50	72	25	$\chi^2_{15:1}=0.89^{ns}$, $\chi^2_{9:3:3:1}=4.79^{ns}$
IR62829A/Poya	49.65 ± 2.11	285	85	63	73	64	$\chi^2_{3:1}=0.98^{ns}$, $\chi^2_{1:2:1}=3.68^{ns}$
IR68899A/Amol-2	70.41 ± 2.30	272	142	59	56	15	$\chi^2_{15:1}=0.25^{ns}$, $\chi^2_{9:3:3:1}=2.76^{ns}$
IR68899A/IR50	68.10 ± 1.46	328	169	74	66	19	$\chi^2_{15:1}=0.12^{ns}$, $\chi^2_{9:3:3:1}=4.28^{ns}$
IR68899A/Poya	47.20 ± 1.90	568	149	123	135	161	$\chi^2_{3:1}=3.38^{ns}$, $\chi^2_{1:2:1}=5.26^{ns}$

F (fertile) = >80% Spikelet fertility; PF (partially fertile) = 60 - 85%; PS (partially sterile) = 1 - 60%; CS (completely sterile) = 0.

Table 4. Pollen fertility of plants in testcross populations.

Test cross	Number of plants					Segregation ratio	Number of Rf gene
	Total	F	PF	PS	CS		
IR58025A//(IR58025A/Amol-2)	111	33	29	25	24	$\chi^2_{1:1:1:1}=2.47^{ns}$	2
IR58025A//(IR58025A/IR50)	135	30	40	33	32	$\chi^2_{1:1:1:1}=1.68^{ns}$	2
IR58025A//(IR58025A/Poya)	119	25	17	8	69	$\chi^2_{1:1}=3.03^{ns}$	1
IR62829A//(IR62829A/Amol-2)	122	35	31	24	32	$\chi^2_{1:1:1:1}=2.12^{ns}$	2
IR62829A//(IR62829A/IR50)	107	28	35	23	21	$\chi^2_{1:1:1:1}=4.35^{ns}$	2
IR62829A//(IR62829A/Poya)	127	30	16	10	71	$\chi^2_{1:1}=1.77^{ns}$	1
IR68899A//(IR68899A/Amol-2)	130	41	32	22	35	$\chi^2_{1:1:1:1}=5.81^{ns}$	2
IR68899A//(IR68899A/IR50)	115	33	24	31	27	$\chi^2_{1:1:1:1}=1.68^{ns}$	2
IR68899A//(IR68899A/Poya)	148	30	26	7	85	$\chi^2_{1:1}=3.27^{ns}$	1

F (fertile) = > 70% Pollen fertility; PF (partially fertile) = 50 - 70%; PS (partially sterile) = 1 - 50%; CS (completely sterile) = 0.

IR58025A//IR58025A/Amol-2, IR58025A//IR58025A/IR50, IR62829A//IR62829A/Amol-2, IR62829A//IR62829A/IR50, IR68899A//IR68899A/Amol-2 and IR68899A//IR68899A/IR50 (Tables 4 and 5). These testcross progenies segregated in the expected ratio of 1 fertile: 1 partially sterile: 1 partially fertile: 1 completely sterile plants which is expected when two independently segregating dominant genes with additive effects, one of them having stronger effect, control fertility restoration of WA cytoplasm in rice varieties Amol-2 and IR50. Such that if both genes are present, fertility are like the restorer variety Amol-2 and IR50; if the gene with stronger fertility restoration ability is present alone fertility is somewhat reduced (partial fertility ranging between 50 - 70% for pollen and 60 -80% for spikelet), but if the gene with weaker restoration ability is present alone, plants show partial sterility ranging between 1 - 50% (for pollen) and 1-60% (for spikelet). The plants possessing the double recessive genotype are completely sterile like IR58025A, IR62829A and IR68899A. These results confirmed the earlier findings of Yao et al. (1997), Jing et al. (2001) and Sheeba et al. (2009), who reported that two independently segregating dominant genes control fertility restoration of WA cytoplasmic male sterility system in rice.

Table 5. Spikelet fertility of plants in testcross populations.

Test cross	Spikelet fertility (%) Mean ± Se	Number of plants					Segregation ratio
		Total	F	PF	PS	CS	
IR58025A//(IR58025A/Amol-2)	52.97 ± 2.01	111	38	25	22	26	$\chi^2_{1:1:1:1} = 5.35^{ns}$
IR58025A//(IR58025A/IR50)	51.60 ± 0.92	135	43	31	29	32	$\chi^2_{1:1:1:1} = 3.51^{ns}$
IR58025A//(IR58025A/Poya)	33.02 ± 0.54	119	34	10	5	70	$\chi^2_{1:1} = 3.70^{ns}$
IR62829A//(IR62829A/Amol-2)	50.36 ± 1.80	122	39	27	23	33	$\chi^2_{1:1:1:1} = 4.81^{ns}$
IR62829A//(IR62829A/IR50)	54.11 ± 0.92	107	37	25	22	23	$\chi^2_{1:1:1:1} = 5.39^{ns}$
IR62829A//(IR62829A/Poya)	32.97 ± 0.76	127	33	14	7	73	$\chi^2_{1:1} = 2.84^{ns}$
IR68899A//(IR68899A/Amol-2)	51.64 ± 1.17	130	42	32	21	35	$\chi^2_{1:1:1:1} = 7.03^{ns}$
IR68899A//(IR68899A/IR50)	50.08 ± 2.11	115	36	25	24	30	$\chi^2_{1:1:1:1} = 2.54^{ns}$
IR68899A//(IR68899A/Poya)	32.92 ± 1.07	148	35	21	7	85	$\chi^2_{1:1} = 3.27^{ns}$

F (fertile) = >80% Spikelet fertility; PF (partially fertile) = 60 - 85%; PS (partially sterile) = 1 - 60%; CS (completely sterile) = 0.

Table 6. Analysis of variance for combining ability of pollen and spikelet fertility in rice.

Sources of variation	df	MS	
		Spikelet fertility	Pollen fertility
Replication	2	0.153	0.035
Genotypes	14	4135.81 **	3636.26 **
Parents (p)	5	7327.58 **	6267.26 **
P vs C	1	20861.31 **	18203.04 **
Crosses (c)	8	50.25 **	166.05 **
Lines	2	1.802 *	131.275 **
Testers	2	182.01 **	141.275 **
L x T	4	8.61 **	205.89 **
Error	28	0.394	1.854

* and **; Significant at p = 0.05 and p = 0.01 levels, respectively, based on F-test.

The F2 population of IR58025A/Poya, IR62829A/Poya and IR68899A/Poya crosses segregated in the ratio of 3 pollen fertile: 1 completely sterile plants ($\chi^2 = 1.24$, $\chi^2 = 0.98$ and $\chi^2 = 3.38$, respectively) which is expected when one dominant gene control fertility restoring ability of Poya variety (Tables 2 and 3). This finding was also confirmed by the segregation pattern of the plants for spikelet fertility in the F2 population of crosses and the testcross progeny of IR58025A// IR58025A/Poya, IR62829A//IR62829A/Poya and IR68899A//IR68899A/Poya (Tables 3, 4 and 5). The testcross progeny exhibited segregation ratio of 1 fertile: 1 completely sterile plant ($\chi^2 = 3.03$, $\chi^2 = 1.77$ and $\chi^2 = 3.27$, respectively), thereby indicating presence of a major fertility restorer locus in the restorer variety Poya. For the restorer variety Poya, the plants having dominant allele in homozygous or heterozygous condition (R1-) will be fertile. The plants having homozygous recessive alleles (r1r1) will be completely sterile. These results confirmed by Anandakumar and Subramaniam (1992), and Gyan et al. (2003), reported that cross IR58025A/PRR-78 showed a segregation ratio of 3:1, confirming the monogenic inheritance of fertility restoration observed in this cross.

Combining ability of fertility restoration

Analysis of variance of combining ability for pollen and spikelet fertility revealed significant differences among genotypes, crosses, lines, testers and line x tester interactions (Table 6). The significant differences among

Table 7. Variances of GCA and SCA and proportional contribution of lines, testers and their interactions to total variance for pollen and spikelet fertility in rice.

Source	Spikelet fertility	Pollen fertility	Source	Spikelet fertility	Pollen fertility
δ^2_{gca}	2.31	1.93	Due to lines	0.89	19.18
δ^2_{sca}	2.74	68.01	Due to testers	90.536	20.64
$\delta^2_{gca} / \delta^2_{sca}$	0.84	0.028	Due to lines x testers	8.57	60.18

Table 8. GCA effects of parents for pollen and spikelet fertility in rice.

Line	GCA		Tester	GCA	
	Spikelet fertility	Pollen fertility		Spikelet fertility	Pollen fertility
IR58025 A	0.26	-3.99 **	Amol-2	3.35 **	3.36 **
IR62829 A	0.25	3.61 **	IR50	-5.11 **	-4.36 *
IR68899 A	-0.52	0.41	Poya	1.76 *	1.025
SE (Sgi) female	0.2	0.45	SE (Sgi) male	0.2	0.45
SE (Sgi - Sgj)	0.29	0.64	SE (Sgi - Sgj)	0.295	0.64

*, ** Combining ability estimate significantly different from zero at p = 0.05 and 0.01, respectively, based on a T-test.

the lines, testers and lines x testers indicated that the genotypes had wide genetic diversity among themselves. The mean sum of squares due to parents versus crosses also differed significantly for pollen and spikelet fertility. The significance of the means of sum of squares due to lines and testers indicated a prevalence of additive variance. However, means of sum of squares due to line x tester were also significant for pollen and spikelet fertility, indicating the importance of both additive and non-additive variance. The ratio of $\delta^2_{gca} / \delta^2_{sca}$ was less than unity for the pollen and spikelet fertility also indicated pre-dominance of non-additive genetic variance (Table 4). It suggested greater importance of non-additive gene action in its expression and indicated very good prospect for the exploitation of non-additive genetic variation for pollen and spikelet fertility through hybrid breeding. Several workers reported the predominance of dominant gene action for pollen and spikelet fertility percentage (Yao et al., 1997; Gyan et al., 2003), while Hoan et al. (1998) and Kumari et al. (1998) reported the predominance of additive gene action.

The proportional contribution of lines, testers and their interactions showed that line x tester interaction played an important role toward pollen fertility (60.18%), indicating predominant female x male interaction influence for this trait (Table 7). For example, pollen fertility in IR58025A/Poya cross was 70.96%, but in IR62829A/Poya and IR68899A/Poya crosses were 90.22 and 89.40%, respectively. However, testers played an important role toward spikelet fertility (90.53 %), indica-ting predominant testers influence for this trait. An estimation of the general combining ability (GCA) effect of lines and testers revealed that the parents IR62829A and Amol-2 were good combiners for pollen fertility percentage and showed superior gene effect (Table 8). Also Amol-2 and Poya varieties were good combiners for spikelet fertility percentage.

The usefulness of a particular cross in exploiting heterosis is judged by the specific combining ability (SCA) effect. IR58025A/IR50 showed the highest SCA effect (8.55) for pollen fertility, followed by the crosses IR68899A/Poya, IR62829A/Poya and IR68899A/Amol-2 (Table 9). These crosses were derived from parents hav-ing different gene effects. The cross of IR58025/IR50 involved parents with low x low GCA, indicating the involvement of dominant x dominant gene interaction and suggesting that the epistatic gene action may be due to genetic diversity in the form of heterozygous loci. On the other hand, cross of IR68899A/Amol-2 involve parents with high x low GCA effects, indicating involvement of additive x dominant gene interaction. Peng and Virmani (1999) also reported the possibility of interaction between positive alleles from good combiners and negative alleles from poor combiners in high x low cross combinations. Thus, in these crosses, high SCA for pollen fertility were attributed to dominance and epitasis gene action. The hybrids IR58025A/IR50, IR62829A/IR50, IR62829A/Poya and IR68899A/Poya have high heterobeltiosis (Table 9). IR58025A/IR50 showed the highest SCA effect (1.733) for spikelet fertility, followed

Table 9. SCA effects and heterobleltiosis of hybrids.

Cross	SCA effect		Heterobleltiosis (%)	
	Spikelet fertility	Pollen fertility	Spikelet fertility	Pollen fertility
IR58025A/Amol-2	-0.327	0.001	98.07	83.01
IR58025A/IR50	1.733 **	8.55 **	99.56	124.49
IR58025A/Poya	-1.405 **	-8.57 **	96.75	65.81
IR62829A/Amol-2	-0.92 *	-2.97 **	96.78	93.40
IR62829A/IR50	-0.044	-0.14 ns	95.37	121.56
IR62829A/Poya	0.967 *	3.08 **	101.97	110.82
IR68899A/Amol-2	1.25 **	2.94 **	99.78	99.44
IR68899A/IR50	-1.688 **	-8.43 **	89.8	90.34
IR68899A/Poya	0.439	5.47 **	99.112	108.90
SE	0.36	0.79		
SE (Sgi - Sgj)	0.51	1.11		

*, ** Combining ability estimate significantly different from zero at p = 0.05 and p = 0.01, respectively, based on a T-test.

by the crosses IR68899A/Amol-2 and IR62829A/Poya (Table 9). These crosses have spikelet fertility the same as with the male parents. Heterobeltiosis offers a greater scope for exploiting hybrid vigor. In the crosses IR68899A/Poya and IR62829A/Poya which had high and significant SCA (Table 9), and with regard to be monogenic of Poya variety (Tables 2, 3 and 4), increase of pollen fertility in this crosses than the Poya variety was due to over-dominance of gene action.

DISCUSSION

Hybrid rice research now concentrates on the conversion and identification of stable local CMS lines and effective restorers from local elite lines through repeated back-crossing. CMS lines developed from Iran are used to maintain the pace of hybrid rice development. Chances of success are greater if outstanding parents with favorable alleles are chosen which on crossing would give, heterotic hybrids. This study aimed to identify good restorers and CMS lines for heterotic rice breeding. One to two genes appeared to control the restoration. Poya variety restored the fertility of IR58025A, IR62829A and IR68899A monogenically, but Amol-2 and IR50 varieties restored the fertility of CMS lines by two independent major genes. Appearance of partial fertile segregants in crosses with complete restorers suggested the probable role of modifiers in fertility restoration.

The results discussed above revealed that except Poya which has one dominant restorer gene showing in F2 and testcross progenies, male fertility restoration for WA cytoplasm in Amol-2 and IR50 is controlled by two dominant genes which segregate independent of each other and have additive effects. One of the two genes appears to be stronger than the other in fertility restoration of WA cytoplasm. These results suggested that as compared to restorer lines with single dominant genes (for example Poya), use of restorer lines carrying two dominant genes for fertility restoration should be preferred to develop hybrid rice varieties by using CMS lines with WA cytoplasm imparting male sterility because such F1 hybrids are expected to have high fertility and will consequently give more yield (Guha Sarkar et al., 2002; Li et al., 2007). We are, therefore, using these restorer varieties with various CMS lines possessing WA cytoplasm to develop high yielding F1 hybrid varieties of rice. These restorer varieties were used in our rice breeding programme to develop more restorers with diverse genetic background. Studies are also under way to determine allelic relationships among the restorer genes of Amol-2, IR50 and Poya.

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