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Research Note

Identification of restorers and maintainers for development of rice hybrids suitable for aerobic condition in rice (*Oryza sativa* L.)

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Abstract
Two cytoplasmic male sterile (CMS) lines of rice having wild abortive WA and one line with Kalinga cytoplasmic male sterility source were crossed with eleven testers to identify their restorer/maintainer nature. Total of 33 hybrids were subjected to pollen and spikelet fertility analysis along with their parents. Most of the genotypes expressed differential fertility reactions when crossed with CMS lines. Testers were identifying as potential restorers. None of the genotypes were identified as potential maintainer based on pollen and spikelet fertility. Among testers only Kranti was identified as potential restorer for all the 3 CMS lines whereas, Danteshwari was identified as potential restorer for 2 CMS lines IR 58025A and IR 79156A. The other testers *viz.*, MTU 1010, Abhaya, Indira Barani Dhan 1, IR 681444, Chandrahasini, were identified as potential restorer for all three CMS lines. Tester Pusa Basmati was identified as partial maintainer for all the three CMS lines. Potential restorers identified in the present investigation could be used to develop good, high yielding and promising rice hybrids with other genotypes.

Key words
Rice, CMS lines, restorers, maintainers

Heterosis breeding in rice is valuable only when promising restorers are identified for different sources of cytoplasmic-genic male sterile (CMS) lines (Pradhan *et al*., 1992). The use of cytoplasmic genetic male sterility system in developing hybrids in crops is possible only when effective restorers are identified (Sharma *et al.*, 2012). A number of cytosterile maintainers and restorer lines in rice have been developed to diversify the genetic and cytoplasmic base of commercial F_1 rice hybrids. The CMS lines developed outside the country are being used as such in developing rice hybrids in India as well as in the state. Therefore, it is essential to identify locally adapted maintainers and restorers among the lines developed using conventional breeding procedures, which could be converted into CMS lines for wide adaptability (Jayasudha and Sharma, 2010).

Exotic CMS line IR 58025A and IR79156A have been widely used in 3-line breeding system and therefore, incorporation of indigenous CMS lines *viz*., CRMS 32A and CRMS 31A have also enormous value to develop locally adopted rice hybrid. Pollen and spikelet fertility or both have been used as an index to fix restoration ability of the genotypes (Sutaryo, 1989). The percentage of fertile pollens is the most reliable criterion for evaluating pollen fertility (Veeresha *et al.* 2013). Considering the importance of heterosis breeding the present investigation was undertaken with the objective to identify different restorers and maintainers for three CMS lines from the local and high yielding rice (*Oryza sativa* L.) genotypes.

The experimental material comprised of three CMS lines from two different CMS sources *viz.*, IR 58025A and IR 79156A (Wild Abortive) and CRMS 32A (Kalinga) and eleven genetically diverse testers- MTU 1010, Abhaya, RHZ-2, Danteshwari, Chapti-gurmatiya, Chandrahasini, IR 681444, Pusa basmati, Dukong, Kranti, Indira Barani Dhan 1. A set of 33 hybrids were generated in Line x Tester fashion (Kempthrone, 1957) for the purpose and evaluated along with their corresponding parents in Randomized Complete Block Design with three replications at research cum instructional farm, Department of Genetics and Plant Breeding, IGKV Raipur, Chhattisgarh during *kharif* 2015. Twenty-one days old seedlings of 33 hybrids and 11 parents were transplanted in the field. A standard spacing of 20 x 20 cm was adopted for planting and 10 plants were maintained in a single row. Single seedling per hill was transplanted. Recommended package of practices were adopted.

Pollen studies were carried out at flowering time to assess fertility / sterility status of F_1 plants. For this purpose, 15-20 spikelets from the just emerged panicles of five randomly selected plants were collected in a vial containing 70% ethanol. All the anthers from at least three to four spikelets were taken out with the help of forceps and placed on a glass slide with a drop of distill water. The anthers were gently crushed by using a needle to release the pollen grains. Then the pollen grains were stained with one percent iodine potassium iodide (I-KI) solution. After removing the debris, a cover slip was placed and the slide was observed under the microscope and the pollen fertility (%) was estimated.

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Five panicles of each test cross were covered with butter paper bags to avoid foreign pollen contamination and were harvested at maturity for estimation of spikelet fertility/sterility. The well filled and chaffy spikelets of each panicle were counted and spikelet fertility (%) was estimated. The following formulae were used to calculate pollen sterility (%) and spikelet fertility (%).

$$\text{Pollen sterility} = \frac{\text{No.of sterile pollens}}{\text{Total No.of pollens}} \times 100$$

$$\text{Spikelet fertility} = \frac{\text{Filled grains per panicle}}{\text{Total spikelets per panicle}} \times 100$$

Classification of parental lines as restorers and maintainers is done on the basis of method proposed by (Virmani *et al.* 1997). According to this method the genotypes that could produce 0-1 per cent pollen fertility and 0.0-0.1 per cent spikelet fertility were classified as maintainers, 1.1-50 per cent pollen fertility and 0.1-50 per cent spikelet fertility were classified as partial maintainers, 50.1-80 per cent pollen fertility and 50.1-75 per cent spikelet fertility were classified as partial restorers and >80 per cent pollen fertility and >75 per cent spikelet fertility were classified as potential restorers.

The restorers and maintainers identified in the present investigation are presented in the Table 1. The studies on pollen and spikelet fertility percentages indicated that none of the hybrids possessed complete pollen and spikelet sterility. So, none of the hybrids could be identified as maintainer, of course partial maintainers were identified. Dukong and Pusa basmati have been identified as partial maintainers for the CMS line IR 58025A.

The parents Dukong, Pusa basmati have been identified as partial maintainers for the line IR79156A. The genotypes RHZ-2, Pusa basmati have been identified as partial maintainers in relation to line CRMS 32A. The lines identified as partial maintainers can be further multiplied and back crossed with their respective F1's to look for completely sterile back cross progenies so that these can be developed as new CMS lines. Similar work plan was reported by Singh and Singh (2000) and Durai and Nadarajan (2007). The parents MTU 1010, Kranti, IR 681444, Danteshwari, can be considered as potential restorers for the CMS line IR 58025A. Parents Kranti, Danteshwari were identified as potential restorers for the CMS line IR 79156A. Whereas the parents, Indira Barani Dhan 1, Kranti, Chandrahasini, Abhaya can be considered as potential restorers for CMS line CRMS 32A. MTU 1010, Indira Barani Dhan 1, IR 681444 Abhaya, and Chandrahasini have been considered as potential restorer for all the three CMS lines. Hence these can be used as potential restorer to develop high yielding, aerobic rice hybrids. Hybrid CRMS 32A/Chapti-gurmatiya showed highest pollen fertility (91.85%) and spikelet fertility (47.69%) followed by crosses IR 79156A/Kranti (89.39% and 62.97%), CRMS 32A/MTU 1010 (88.60% and 76.90%), CRMS 32A/Abhaya (88.23% and 81.63%), IR 58025A/MTU 1010 (87.64% and 51.01%), IR 58025A/Indira Barani Dhan 1 (87.64% and 65.35%), CRMS 32A/ Indira Barani Dhan 1 (86.94% and 52.59%), CRMS 32A/Kranti (86.28% and 48.09%), IR 79156A/Chapti-gurmatiya (85.85% and 2.42%), IR 58025A/RHZ-2 (84.71% and 55.89 %) IR 58025A/IR 691444 (84.65% and 92.19%), IR 58025A/Danteshwari (82.58% and 84.94%), IR 58025A/ Kranti (82.58% and 49.20%). Therefore, these crosses can be effectively utilized as good restorer lines to develop high yielding rice hybrids.

In some cases, the same genotype behaved as a restorer for one CMS line and as partial maintainer or partial restorer for the other CMS line. Similar type of results was reported by Bisne and Motiramani (2005).

The variations in behaviour of fertility restoration indicate that either the fertility-restoring genes are different or that their penetrance and expressivity varied with the genotypes of the parents or the modifiers of female background. This could be due to differential nuclear cytoplasmic interactions between the testers and CMS lines. This kind of the differential reaction of the same genotype in restoring the fertility of different CMS lines of same cytoplasmic source was reported by Hariprasanna *et al*. (2005), Murugan and Ganesan (2006) and Jayasudha and Sharma (2010). The investigation concludes seven genotypes MTU 1010, Abhaya, Indira Barani Dhan 1, IR 681444, Chandrahasini, Kranti and Danteshwari were identified as potential restorers, among which only one genotypes *viz*., Kranti was identified as potential restorer for all three CMS lines (IR58025A, IR79156A and CRMS 32A). Potential restorers identified in the present study can be further used for developing good, high yielding rice hybrids.

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Table 1. Identified Restorers and Maintainers (Based on pollen and spikelet fertility)

S. No.	Crosses	Pollen Fertility		Spikelet Fertility		Based on both
		%	Class	%	Class	
1	IR-58025A/MTU1010	87.64	PR*	50.17	PR	PR*/PR
2	IR-58025A/Abhaya	89.94	PR*	62.50	PR	PR*/PR
3	IR-58025A/Indira Barani Dhan 1	87.61	PR*	65.35	PR	PR*/PR
4	IR-58025A/Chandrahasini	79.82	PR	41.39	PM	PR/PM
5	IR-58025A/RHZ 2	84.71	PR*	55.89	PR	PR*/PR
6	IR-58025A/Dukong	36.01	PM	23.09	PM	PM
7	IR-58025A/Kranti	82.58	PR*	49.20	PM	PR*/PR
8	IR-58025A/Danteshwari	82.58	PR*	84.94	PR*	PR*
9	IR-58025A/Chapti-gurmatiya	68.05	PR	74.58	PR	PR
10	IR-58025A/IR 681444	84.65	PR*	92.19	PR*	PR*
11	IR-58025A/Pusa Basmati	6.24	PM	2.11	PM	PM
12	IR-79156A/MTU1010	66.28	PR	60.40	PR	PR
13	IR-79156A/Abhaya	60.44	PR	66.82	PR	PR
14	IR-79156A/IBD 1	65.22	PR	39.55	PM	PR/PM
15	IR-79156A/Chandrahasini	72.24	PR	58.44	PR	PR
16	IR-79156A/RHZ 2	61.90	PR	55.85	PR	PR
17	IR-79156A/Dukong	5.02	PM	17.31	PM	PM
18	IR-79156A/Kranti	89.39	PR*	62.97	PR	PR*/PR
19	IR-79156A/Danteshwari	78.08	PR	66.22	PR	PR
20	IR-79156A/Chapti-gurmatiya	85.85	PR*	2.42	PM	PR*/PM
21	IR-79156A/IR681444	57.49	PR	88.17	PR*	PR/R
22	IR-79156A/Pusa Basmati	5.33	PM	1.73	PM	PM
23	CRMS-32A/MTU 1010	88.60	PR*	51.01	PR	PR*/PR
24	CRMS-32A/Abhaya	88.23	PR*	81.63	PR*	PR*
25	CRMS-32A/Indira Barani Dhan 1	86.94	PR*	52.59	PR	R/PR
26	CRMS-32A/Chandrahasini	61.09	PR	79.57	PR	PR
27	CRMS-32A/RHZ 2	45.29	PM	46.78	PM	PM
28	CRMS-32A/Dukong	74.65	PR	91.36	PR*	PR*/PR
29	CRMS-32A/Kranti	86.28	PR*	48.08	PM	PR*/PM
30	CRMS-32A/Danteshwari	78.01	PR	80.63	PR*	PR/PR*
31	CRMS-32A/Chapti-gurmatiya	91.85	PR*	47.69	PM	PR*/PM
32	CRMS-32A/IR 681444	59.95	PR	40.48	PM	PR/PM
33	CRMS-32A/Pusa Basmati	4.28	PM	7.18	PM	PM

PM – Partial Maintainer, PR- Partial Restorer, PR*-potential Restorer, M- Maintainer