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Suitability of Maintainers and Restorers for CMS Lines in Rice

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Hybrid rice technology has been successfully developed and is one of the potent options for increasing rice production. The use of cytoplasmic genetic male sterility system in developing hybrids in crops is possible only when effective restorers are identified. Pollen (or) spikelet fertility or both have been used as an index to fix the restoration ability of the lines. In the present investigation 19 genotypes were crossed with four stable CMS lines during Rabi 2009-2010 at Paddy Breeding Station, Coimbatore. Genotypes were categorized as effective restorers (> 80 % spikelet fertility), partial restorers (20-79% spikelet fertility), partial maintainers (10-19 % spikelet fertility) and maintainers (<10 % spikelet fertility). A total of eighteen effective restorers and eight maintainers were identified among 100 genotypes against four CMS lines. The pollen fertility per cent of hybrids varied from 0 to 98.30%. The spikelet fertility of F_1s ranged from 0 (IR 80151 A x CB 06 107) to 98.65 (TNAU CMS 2A x CB 06 734). Maximum number of maintainers were observed against IR 80151A. The maximum number of effective restorers were observed against TNAU CMS 2A. The identified restorers and maintainers could be utilized for development of new rice hybrids and CMS lines respectively in future.

Key words: Restorers, maintainers, pollen fertility, spikelet feritility.

Rice is a staple food of India, providing 43% of calorie requirement for more than 70% of Indian population. It occupies a predominant position among major food grains fulfilling about 60 per cent dietary requirement, 20 per cent of calorie and 14 per cent protein requirement of the world population. To meet the demands of increasing population and maintain this self-sufficiency, the present production level of around 90 million tons, needs to be increased up to 120 million tons by the year 2020. Hybrid rice is one of the practically feasible and readily adoptable technologies to increase production and productivity of rice in India and hence it has been included as an important component under the National Food Security Mission (NFSM). The availability of stable cytoplasmic male sterility and fertility restoration system is vital for commercial exploitation of heterosis in any crop. With the discovery of the wild abortive (WA) male sterility inducing cytoplasm from *Oryza sativa spontanea* and subsequent development of three line hybrids made a breakthrough in exploitation of heterosis in rice (Lin and Yuan, 1980). The use of cytoplasmic genetic male sterility system in developing hybrids in crops is possible only when effective restorers are identified. The CMS lines introduced from China are unstable to use as such in developing hybrid rice in India. Therefore, it is imperative to identify maintainers and restorers among the lines developed through conventional breeding procedures. In present investigation, efforts were made to identify the maintainers and restorers for four CMS lines to develop potential rice hybrids.

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Materials and Methods

The 19 genotypes were crossed with four cytoplasmic male sterile lines *viz.*, CRMS 31A, CRMS 32A, IR 80151A and TNAUCMS 2A during Rabi 2009-10. Crossing was done by adopting clipping method. In the CGMS lines individual plants with complete pollen sterility was identified by observing the pollen grains under the microscope using one per cent Iodine potassium iodide stain. The spikelets were clipped off one third from the top without damaging the stigma, between 7.00 and 9.30 a.m. Immediately after clipping, the panicles were covered with butter paper covers. At the time of anthesis, panicles with fully opened spikelets were collected from the male parents and the pollen grains were dusted over the clipped panicles between 10.30 and 11.30 am. Crosses were effected between female and male parents and a total of 76 cross combinations were obtained. After 25 days, the matured panicles were harvested. The F_1 generations of all the crosses were raised during *kharif* 2010 in a test cross nursery along with their respective parents. Each entry was planted with a

spacing of 20 x 20 cm with two replications. Identification of maintainers and restorers was carried out by observing pollen and spikelet fertility. Potential maintainers were identified as having >90% pollen sterility and <10% spikelet fertility and effective restorers as having <10% pollen sterility and >90% spikelet fertility.

The pollen fertility was calculated as follows

In hybrid evaluation, all the 76 hybrids were

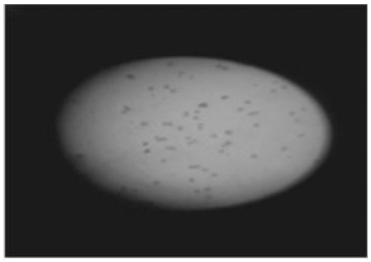
$$\frac{\text{Number of (unstained withered + unstained spherical + partly stained round) pollen}}{\text{Total number of pollen counted (including fertile)}} \times 100$$

studied for spikelet fertility per cent by bagging the primary panicles at booting to heading stage before anthesis. On the basis of spikelet fertility, pollen parents were classified as effective restorer (> 80 % spikelet fertility), partial restorers (20 to 80 % fertility), partial maintainers (5 to 20 % fertility) and effective maintainers (< 5 % fertility). The proportion of number of fully developed grains to the total number of spikelets was calculated as spikelet fertility.

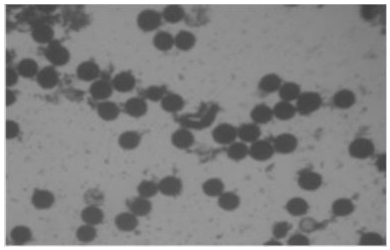
$$\text{Spikelet fertility (\%)} = \frac{\text{Number of filled grains per panicle}}{\text{Total number of spikelets per panicle}} \times 100$$

Results and Discussion

The genotypes were identified as restorers and maintainers as per their fertility restoring and sterility maintaining ability in F1 plants of respective CMS lines.The pollen fertility per cent of hybrids was varying from 0.0 (IR 80151A x CB 09 174) to 98.30 % (TNAU CMS 2A x CB 08 529). A very low magnitude of pollen and spikelet fertility was observed for hybrids (Table 1). The lines identified as effective maintainers can be further 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.



IR 80151 A x CB 06 107 (M)



TNAU CMS 2A x CB 09 105 (R)

Fig. 1. Observation on pollen fertility studies

Five genotypes *viz.,* CB 09 149, CB 09 194, CB 09 174 and CB 06 107 were observed as maintainers for IR 80151A line with spikelet fertility below 10 % in F1s (Fig.1). Two genotype namely CB 09 165 and IET 20601 showed above 80 % spikelet fertility and these were identified as potential restorers for development of new hybrid

Table 1. Pollen and spikelet fertility (%) in F1's and identification of restorers and maintainers for various CMS lines

CMS lines	Restorers	Maintainers
CRMS 31A	CB 08 144, DRR 3306, CB 09 169, CB 09 153, CB 07 115, CB 09 172, CB 09 165	-
CRMS 32 A	DRR 3306, CB 09 165, IET 27044, CB 09 169, CB 09 156	CB 09180, CB 07115
IR 80151 A	CB 09 165, DRR 3306, IET 20601	CB 09149, CB 09194, CB 09174, CB 06107
TNAU CMS 2 A	CB 09 105, Kanto 5, DRR 3306, IET 20601, CB 09 106, CB 06 102, CB 06 563, IET 20427, CB 08 529, CB 06 734, CB 09 165	CB 09107

combinations. Ali and Khan (1995) and Ingale *et al* (2008) reported that RTN 40-3-1-1, IR 47456 and PK 4112 were potential maintainers and PK 4029-2 and PK 4029-3 were restorers for IR 58025A and IR 62829A respectively in their studies.

In some cases, the same genotype behaved as a restorer for one CMS line and as a partial restorer for the other CMS line. Tester CB 07 115 behaved as an effective restorer for CMS line CRMS 31A and was found to be partial restorer for CRMS 32 A. Tester DRR 3302 was a partial restorer for CRMS 31 A and was found to be Maintainer for CRMS 32 A. Similar results were reported by other workers too most of the genotypes expressed differential fertility reactions when crossed with CMS tries ... WA cytoplasm. The pollen and spikelet fertility were

highly influenced by environmental conditions. (Ingale et al, 2008). This might be due to different source of CMS, because all other CMS source used are of 'WA' source but CRMS 32A is a 'Kalinga' source. Source of CMS line also played major role in restoring ability of the CMS lines. The tester Kanto 5 behaved as partial maintainer for IR 80151A and was found to be effective restorer for TNAU CMS2A. 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 kind of 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) and Murugan and Ganesan (2006). This could be due to differential nuclear cytoplasmic interactions between the testers and CMS lines. The spikelet fertility of F_1s ranged from (IR 80151 A x CB 06 107) to 94.70 (CRMS 31 A x DRR 3306).

Table 2. Proportion of maintainers, partial maintainers/partial restorers and restorers in crosses of four CMS lines of rice

Name of the CMS line	Total crosses	No. of maintainers observed	Number of partial maintainer/partial restorer observed	No. of restorers observed	Proportion of M:PM/ PR:R in percent
CRMS 31 A	12	-	5	7	0.00:41.66:58.33
CRMS 32 A	28	3	20	5	10.71:71.43:17.86
IR 80151 A	13	4	5	4	30.76:38.46:30.76
TNAU CMS 2A	23	1	12	10	04.35:52.17:43.48
Total	76	8	42	26	10.5:55.3:34.3

The proportion of maintainers, partial maintainers/partial restorers and restorers were recorded in four CMS lines (Table 2). The maximum proportions of maintainers were observed for IR 80151 A (30.76%) followed by CRMS 32 A (10.71%) and TNAU CMS 2A (4.35%), which could be used conversion of new CMS lines in hybrid rice breeding programs. The maximum proportions of restorers were observed for CRMS 31 A (58.33%), followed by TNAU CMS 2A (43.48%), IR 80151A (30.76%) and CRMS 32 A (17.86%) which could be used for development of new rice hybrid combinations**.** The proportion of 45.82% maintainers, 203.72% partial maintainer/restorer and 150.43% restorers were observed in 76 crosses with four CMS lines under study. The potential maintainers for all the CMS lines are being used in the back cross program to develop new CMS line. There were instances were the classification of tester based on the pollen fertility did not correlate with the classification based on the spikelet fertility. The testers DRR 3306 behaved complete restorer for all the four CMS lines. This tester could be utilized in the heterosis breeding after testing their combining ability and heterosis.

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