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Identification of restorers and maintainers for different 'WA' CMS lines in rice (*Oryza sativa* L.)

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Abstract: The present observations revealed that F_1 hybrids produced by crossing 40 different rice genotypes with 2 CMS lines (IR79156A and Pusa 6A) behaved differently with regard to pollen fertility. Out of the 80 F_1 hybrids, 12 were completely sterile and 22 completely fertile. The remaining 46 hybrids expressed varying degrees of fertility, within the range of partial restorers (21) and partial maintainers (25). Genotypes; IET 21519, IET 22218, IET 22228, IET 22202, IET 21542, Sarju- 52, BPT 5204 and MTU-7029 produced higher fertile hybrids and are hence considered as common effective restorers for both the CMS lines. Genotypes IET 20524, RPBIO-226 and HUR-8-1 were found to be effective restorer for only IR79156A whereas; NDR-359, IDR763 and Type-3 were found to be effective restorer for CMS line Pusa 6A. These restorer genotypes may be tested for heterosis for development of new rice hybrids. HUR-105 and Pant Dhan-4 produced sterile hybrids when crossed with IR79156A. Pant Dhan-12 and Vandana produced sterile hybrids when crossed with Pusa 6A. Four genotypes (IET 22237, NDR-97, Nagina-22 and Karahani) produced sterile hybrids when crossed with both the CMS lines, IR79156A and Pusa 6A. These genotypes may be used for development of new male sterile lines.

Key words: CMS lines, fertility restorers, hybrid rice, maintainer, wild abortive

Introduction

Rice is one of the most important staple crops of India. It contributes to total food grain and cereal production of the country to nearly 43% and 46% respectively. In India, rice is being grown in 42.86 million hectare area with production of 104.32 million tonnes and productivity of 24.34 q/ha (Foreign Agriculture Services/USDA, Office of Global analysis, April 2012). The population of India, is still growing at higher rate, therefore to meet the demand of increasing population, the country has to enhance its rice production up to 135 million tons by 2020. Among several genetic options available to increase the yield, hybrid rice technology is one the strongest tools. But, to exploit this technology, male sterility is a prerequisite. Use of CMS lines and restorer lines which are diverged and locally adapted will not only increase the nuclear diversity but would also help to prevent genetic vulnerability due to the use of a single CMS source (Pradhan *et al*., 1992). The restorers and maintainers for WA cytoplasm were reported earlier by Rosamma and Vijayakumar (2005), Sabar *et al*. (2007), Akhter *et al*. (2008), Das *et al.* (2012), Khan *et al.* (2012), Krishnalatha and Sharma (2012), Sharma *et al.* (2012), Soni *et al.* (2012) and Singh *et al.* (2013). The purpose of diversification requires search for new maintainers and restorers. Hence, the present investigation was undertaken with an objective to identify different restorers and maintainers for three CMS lines from among the local and high yielding rice genotypes.

Materials and Methods

The experimental material consisting of 80 F_1 hybrids and their parental lines and 2 CMS lines (IR79156A and Pusa6A) were grown in single row of 4.0 m with three replications in Randomized Block Design with spacing of 20 x 15 cm during *Rabi* 2012-2013 at Central Rice Research Institute (CRRI), Cuttack, Odisha, India. The investigation was undertaken to estimate pollen and spikelet fertility to identify restorers and maintainers. The purpose of identification of maintainers was to isolate locally adopted genotypes which may be converted into new CMS lines through repeated backcross breeding, while the purpose of identification of restorers was to find out suitable fertility restorers among the locally adapted varieties for the existing CMS lines as a short term strategy for hybrid breeding programme. The pollen and spikelet fertility percentage were used as the fertility index Babu *et. al.* (2010).

For pollen fertility, spikelets were collected from the panicle at the flowering stage. Mature anthers from 5 randomly selected spikelets were squashed, smeared and stained with 1% Iodine Potassium Iodide (IKI) solution and examined under light microscope. Pollen grains were counted at three different spots in the microscopic field. Stained, well-filled and round pollen grains were counted as fertile (viable), while unstained, shriveled and empty pollen grains were considered as sterile (non viable). Pollen viability was calculated and expressed in percentage as:

Table - 1: Per cent pollen and spikelet fertility of 40 genotypes involving two cytoplasmic male sterile lines

Genotypes	IR 79156A		Pusa 6A	
	PF	SF	PF	SF
IET – 20924	68.81	65.88	32.18	25.87
IET – 21519	85.71	84.41	84.73	77.57
IET – 22218	86.77	83.90	87.08	82.74
IET – 22251	52.33	43.33	72.17	65.62
IET – 20935	42.88	33.23	16.17	7.54
IET – 20556	16.96	12.58	24.10	10.61
IET – 22228	84.60	76.41	83.78	76.70
IET – 22225	78.10	76.88	64.17	50.71
IET – 22202	84.66	79.04	85.96	79.72
IET – 21528	46.56	43.90	63.98	53.87
IET – 22237	0.00	0.00	0.00	0.00
IET – 20524	84.43	81.86	67.04	62.15
IET – 21542	87.07	82.12	91.38	83.70
Vardhan	43.33	33.76	71.00	64.96
Akshaya Dhan	33.80	25.06	52.71	51.97
HUR - 3022	68.01	65.33	64.15	49.57
HUR - 105	2.60	0.79	26.20	10.71
HUBR - 2-1	44.46	37.30	66.46	54.56
Rajendra Kasturi	48.00	37.36	36.95	31.73
Sarju- 52	87.48	79.21	91.11	84.06
NDR - 359	34.86	27.10	92.34	84.93
NDR - 97	0.00	0.00	0.00	0.00
Pusa - 1460	73.46	66.90	70.66	59.24
Pant Dhan- 4	0.00	0.00	63.99	58.58
Pant Dhan -12	42.93	39.87	0.00	0.00
Pant SugandhDhan-17	25.11	12.60	71.21	62.81
Khutadhan	29.10	18.89	28.41	17.24
Local Nagina - 22	0.00	0.00	0.00	0.00
Local Karahani	0.00	0.00	0.00	0.00
RPBIO-226	83.28	79.55	68.20	58.47
HUR - 8-1	84.63	85.86	21.12	16.17
BPT - 5204	91.44	82.90	82.35	78.48
Adam Chini	66.31	65.60	57.90	51.70
HUR - 5-2	33.13	26.40	49.83	34.87
Anjali	47.08	43.72	60.37	54.31
IDR - 763	47.41	35.56	85.50	78.73
Type - 3	72.27	66.16	87.14	80.48
MTU - 7029	85.64	78.59	86.34	85.60
Vandana	43.22	32.22	0.00	0.00
Danteswari	62.91	56.28	46.63	50.45

PF = Pollen fertility (%); SF = Spikelet fertility

Table - 2: Fertility classification of 40 Genotypes for two cytoplasmic male sterile (CMS-WA) lines

Genotypes	IR 79156A	Pusa 6A
IET – 20924	PR	PM
IET – 21519	R	R
IET – 22218	R	R
IET – 22251	PM	PR
IET – 20935	PM	PM
IET – 20556	PM	PM
IET – 22228	R	R
IET – 22225	PR	PR
IET – 22202	R	R
IET – 21528	M	PR
IET – 22237	M	M
IET – 20524	R	PR
IET – 21542	R	R
Vardhan	PM	PR
Akshaya Dhan	PM	PR
HUR – 3022	PR	PR
HUR – 105	M	PM
HUBR - 2-1	PM	PR
Rajendra Kasturi	PM	PM
Sarju- 52	R	R
NDR - 359	PM	R
NDR – 97	M	M
Pusa – 1460	PR	PR
Pant Dhan- 4	M	PR
Pant Dhan -12	PM	M
Pant SugandhDhan-17	PM	PR
Khutadhan	PM	PM
Local Nagina– 22	M	M
Local Karahani	M	M
RPBIO-226	R	PR
HUR - 8-1	R	PM
BPT – 5204	R	R
Adam Chini	PR	PR
HUR - 5-2	PM	PM
An Ali	PM	PR
IDR – 763	PM	R
Type – 3	PR	R
MTU - 7029	R	R
Vandana	PM	M
Danteswari	PR	PM

M = Maintainer; R = Restorer; PM = Partial Maintainer; PR = Partial Restorer

$$\text{Pollen fertility (\%)} = \frac{\text{Number of stained pollen grains}}{\text{Number of total pollen grains}} \times 100$$

For spikelet fertility, well-developed filled grains were counted from 5 randomly selected panicles for each test entry in each replication and expressed in percentage. The following formula was used for computing fertility percentage:

$$\text{Spikelet fertility (\%)} = \frac{\text{Number of filled spikelets}}{\text{Number of total spikelets}} \times 100$$

Results and Discussion

Out of the 80 F_1 hybrids having CMS lines with WA cytoplasm, 22 were completely fertile and 12 completely sterile.

Table - 3: Means of spikelet and pollen fertility of parents (only full restorers) in rice

Genotypes	Pollen fertility (%)	Spikelet fertility (%)
Lines		
IR 79156A	0	0
Pusa 6 A	0	0
Testers		
IET 21519	84.96	82.30
IET 22218	85.83	84.86
IET 22228	85.05	84.93
IET 22202	84.33	84.26
IET 21542	83.07	83.13
Sarju- 52	86.78	86.83
BPT 5204	86.93	84.50
MTU-7029	86.36	86.55
Grand Mean	85.41	84.67
SE (m)	0.559	1.179
CD (5%)	1.42	2.79
CD (1%)	2.09	4.13

Table - 4: Means of spikelet and pollen fertility of F_1 hybrids (only full restorers) in rice

F_1	Pollen fertility (%)	Spikelet fertility (%)
IR 79156A x IET 21519	85.71	84.41
IR 79156A x IET 22218	86.77	83.90
IR 79156A x IET 22228	84.60	76.41
IR 79156A x IET 22202	84.66	79.04
IR 79156A x IET 21542	87.07	82.12
IR 79156A x Sarju- 52	87.48	79.21
IR 79156A x BPT 5204	91.44	82.9
IR 79156A x MTU-7029	85.64	78.59
Pusa 6A x IET 21519	84.73	77.57
Pusa 6A x IET 22218	87.08	82.74
Pusa 6A x IET 22228	83.78	76.70
Pusa 6A x IET 22202	85.96	79.72
Pusa 6A x IET 21542	91.38	83.7
Puas 6A x Sarju- 52	91.11	84.06
Puas 6A x BPT 5204	82.35	78.48
Puas 6A x MTU-7029	86.34	85.6
Grand Mean	86.63	80.94
SE(m)	0.765	1.006
CD (5%)	1.64	2.14
CD (1%)	2.25	2.97

The remaining 46 hybrids expressed varying degrees of fertility. Twenty-one of them were partial restorer and the remaining 25 were partial maintainer (Table-1 to 4).

Total eight genotypes *viz.,* IET 21519, IET 22218, IET 22228, IET 22202, IET 21542, Sarju-52, BPT 5204 and MTU-7029 were found to be common effective restorers for CMS lines (IR 79156A and Pusa6A). Genotypes IET 20524, RPBIO-226 and HUR-8-1 were found to be effective restorer for CMS line IR79156A. NDR-359, IDR763 and Type-3 were found to be effective restorer for CMS line Pusa 6A. These restorer genotypes may be tested for heterosis for development of new rice hybrids. HUR-105 and Pant Dhan-4 produced sterile hybrids when crossed with IR79156A. Pant Dhan-12 and Vandana produced sterile hybrids when crossed with Pusa 6A. Four genotypes (IET 22237, NDR-97, Nagina-22 and Karahani) produced sterile hybrids when crossed with IR79156A and Pusa 6A both.

The study concluded that the frequency of potential restorers is higher in number than the frequency of maintainers. The similar results were also reported by Rosamma and Vijay Kumar (2005) and Sharma *et al.* (2012) in rice. The pollen as well as spikelet fertility are highly influenced by environmental conditions. Thus a partial restorer in one environment may behave like complete restorer at another environment. Similarly, the partial or weak maintainer lines under the environment favoring pollen sterility may behave like maintainer lines (Virmani *et al.,* 2003). The lines that restore fertility between 30 to 59% can be safely rejected from the breeding programme. The study suggests that the new CMS lines could be developed by using newly identified maintainers (IET 22237, NDR-97, Nagina-22, Karahani, HUR-105, Pant Dhan-4, Pant Dhan-12 and Vandana) as these lines produced full sterile hybrids with CMS lines which could be converted into new CMS lines through repeated backcrossing. New hybrids could be developed by using the identified restorers (IET 21519, IET 22218, IET 22228, IET 22202, IET 21542, Sarju-52, BPT 5204, MTU-7029 IET 20524, RPBIO-226, HUR-8-1, NDR-359, IDR763 and Type-3) as they have produced full fertile hybrids. These results are in conformity with the findings of Gautam and Singh (2004); Rosamma and Vijay Kumar (2005); Sabar *et al.* (2007), Akhter *et al*. (2008), Das *et al.* (2012), Khan *et al.* (2012), Krishnalatha and Sharma (2012), Soni *et al.* (2012) and Singh *et al.* (2013).

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