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Fertility restoration behaviour of rice genotypes for CMS lines under saline-alkali situation

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ABSTRACT

Field experiment comprised of forty rice hybrids derived from two CMS lines (IR 58025 A and NMS 4 A) and twenty testers following L×T mating design, were conducted during wet season 2006 under saline-alkali soil [pH = 9.65, EC (dSm^{-1}) = 2.21, OC = 0.20 %] at Genetics and Plant Breeding Research Farm of N.D.U.A. & T., Kumarganj, Faizabad. Out of twenty rice genotypes screened for their restoration ability, the eight pollen parents viz., IR 61920-3B-22-2-1, IR 70023-4B-R-12-3-1-1-B, NDRK 5095, NDRK 5086, NDRK 5013, CSRC(S) 14-1-4-0, 22-2-B-2-1-1 and 92-H 51-4 were found as partial restorers for both the CMS lines. Six pollen parents viz., NDRK 5056, NDR 9830119, CST 7-1, 21-2-5-B-1-1, IR 64, and PNL 5-8-1-7-21 were found as partial restorers to only for NMS 4 A and same testers were found restorers for IR 58025 A. Out of 20 pollen parent, only six viz., PNL 1-8-5-17-2, NDR 9830148, IR 72048-B-R-2-2-2-1-B, IR 71829-3R-73-1-2-B, NDRK 5094 and Narendra Usar 3 were found as complete restorer to both the CMS lines. These parents are suggested for recombination breeding to obtain desirable/better segregants with high grain yield and wide compatible genes which enhance the rapid hybrid rice seed production for saline-alkali rice growing ecosystem.

Key words: *rice, cytoplasmic male sterility, pollen viability, spikelet fertility, fertility restoration*

Successful development of hybrid rice, using the wild abortive (WA) type of cytoplasmic male sterility (CMS) in China has attracted the attention of rice geneticists throughout the world including India. In fact, the hybrid rice research was initiated in 1964 (Yuan, 1966) and the genetic tools essential for breeding hybrid rice varieties are as the male sterile line (A-line), maintainer line (B-line) and restorer line (R-line) were developed during 1973 (Yuan and Virmani, 1988). The breeding methodology involves the three approaches (a) Three line method or CMS system which is possible and has been found to be most effective genetic tool for developing hybrids, (b) Two line method or PGMS and TGMS system and (c) One line system or apomictic system which would enable farmers to use their own seed for the successive crops without experiencing genetic segregation. Among them, three lines approach is being widely adopted in India and had resulted in the development of more than one and half dozen of rice hybrids. The CGMS is essentially CMS with a provision of fertility restoration by nuclear gene(s). Hence, it is also referred to as CMS system. The role of cytoplasm in causing male sterility in rice was reported back in the fifties and the first usable cytoplasmic male sterility-fertility restoration system in rice was developed by substituting nuclear genes of *Japonica* variety-Taichung 65 into the cytoplasm of the *Indica* variety Chinsurah Boro II (Sampath and Mohanty, 1954). However, this could not be exploited for commercial hybrid seed production probably due to strict self-pollinating nature of the crop. The first commercially usable CMS line was developed in China during 1973 from a spontaneous male sterile plant isolated in a population of the wild rice *Oryza sativa* f. *spontanea* on Hainen Island. Discovery of this source, designated as Wild Abortive "WA" type is considered a landmark in the history of hybrid rice. China released the first hybrid for commercial cultivation in 1976 under the guidance of

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father of hybrid rice Prof. Yuan Long Ping. Subsequently using WA-based CMS lines *viz*., V 20 A, Zhen-shan 97 A, etc., several heterotic rice hybrids were released for commercial cultivation. The yield advantage of 15-20 % over the best pure line varieties (6.5 t ha^{-1} as against 5.4 t ha^{-1}) proved the key factor for wide adoption of the hybrid rice technology. China currently grows hybrid rice in about 18 m ha which produces 17% of the world's paddy with productivity of 6.78 t ha^{-1}. The increased production (20-25 m t $year^{-1}$) from hybrid rice in China has helped in stabilizing global rice prices and therefore, indirectly helped rural and urban poor. Both saline and sodic soils are wide spread in inland areas as that in UP. Rice is a suitable crop to start with for rehabilitation of those lands under flooded conditions because rice is the obvious choice during the soil reclamation. At present in the country an estimated 8.9 m ha of land is affected by salt of which UP share is 1.29 m ha (International Symposium on Rice, 2004). The present investigation was therefore, aimed to isolate the restorers, partial restorers, partial maintainer and maintainer lines for CMS lines (IR 580265 A and NMS 4 A) under such abiotic stress.

MATERIALS AND METHODS

The experimental materials comprised of two CMS lines (IR 580265 A and NMS 4 A) and twenty salinity and alkalinity resistance/tolerance genotypes as testers (males) to produce 40 rice hybrids in line × tester mating fashion during *kharif* 2004-05. Forty rice hybrids were grown in *kharif* 2005-06 and evaluated along with their parents at Genetics and Plant Breeding Research Farm of N.D.U.A. & T., Kumarganj, Faizabad under saline-alkali soil [pH = 9.65, EC (dSm^{-1}) = 2.21, OC = 0.40 %] situation. Thirty days old and single seedling $hill^{-1}$ was taken for transplanting at 20 × 15 cm distance and row length of 3m for each entry. All the recommended cultural practices were adopted to raise a good crop. The stability of each CMS line was tested by scoring the sterile pollen count and counting the spikelet sterility of the individual plant. Spikelets were collected randomly from the panicle at the flowering stage. Mature anthers from 5 randomly selected spikelets were squished, smeared and stained with 1 per cent 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, shrivelled and empty pollen grains were considered as sterile (non viable). Pollen viability/ fertility was calculated and expressed in percentage as:

$$\text{Pollen fertility (\%)} = \frac{\text{Number of stained pollen grain}}{\text{Total number of pollen grains examined}} \times 100$$

From each entry, 5 panicles were randomly selected for filled grain count. The empty spikelets were considered as sterile and spikelets filled with grain as fertile. Spikelet sterility was expressed in percentage. The percentage of spikelet fertility was calculated as given below:

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

Percentage of fertility of hybrids was used as indices of restoring ability of these lines following IRRI method of scoring. (i) The F_1 plants which scored >80% spikelet fertility were considered as restorer and respective pollen parent as restorer lines. (ii) The F_1 plants showing 10-80% spikelet fertility were considered as partial restorers. (iii) The F_1 plants with 1-10% spikelet fertility were considered as partial maintainers, and (iv) F_1 plants with <1% spikelet fertility were considered as maintainers.

RESULTS AND DISCUSSION

During the course of present investigation, 2 CMS lines were evaluated for their relative stability of pollen sterility in *kharif* 2004-05 and 2005-06 at main campus, NDUA&T, Kumarganj, Faizabad. Both of the CMS lines had 'WA' (wild abortive) source of sterile cytoplasm and found stable. The present findings are in close conformity with the earlier observations of Virmani *et al.* (1981), Mishra and Pandey (1993) and Zaibunnisa *et al.* (2002) suggested the pollen sterility is also enhanced due to salt stress.

The effectiveness of hybrid breeding programme mainly rests on the identification, development, maintenance and evaluation of CMS lines (A-lines), their maintainers (B-lines) and effective restorers (R-lines). The method of developing hybrids using A, B and R lines is popularly called as 'Three line

system' of hybrid rice breeding. A number of CMS lines have been developed from different sources but only few of them are being used in commercial hybrid rice programme. This is due to the reason that many of these CMS lines are handicapped with one or the other problem like abnormal flowering behavior, absence of effective restorers and unstable male sterility.

During the course of the present investigation, 40 crosses were made to identify distinct and effective restorers for both CMS lines *viz.,* IR 58025 A and NMS 4 A. Examination of the F_1's have helped to identify the genotype for the target trait. Out of 20 testers, 8 lines behaved like partial restorers (10-80% spikelet fertility) and 6 as restorer (>80% spikelet fertility) for both the CMS lines, whereas, 6 were restorer for CMS line (IR 58025 A) as well as partial restorer for other CMS line (NMS 4 A). None of the pollen parent was found to behave like maintainer or partial maintainer for both the CMS lines.

Table 1. Pollen viability, spikelet fertility and restoration behaviour of pollen parents with CMS line NMS 4A

Pollen parent	Pollen fertility (%)	Spikelet fertility (%)	Restoration behaviour
IR 61920-3B-22-2-1	56.17	49.35	PR
IR 70023-4B-R-12-3-1-1-B	77.37	71.59	PR
NDRK 5095	58.30	36.91	PR
NDRK 5056	78.38	65.43	PR
NDRK 5086	53.53	50.39	PR
NDR 9830119	79.16	69.54	PR
NDRK 5013	34.04	33.04	PR
CST 7-1	48.78	48.63	PR
21-2-5-B-1-1	87.61	70.06	PR
IR 64	62.72	62.22	PR
CSRC(S) 14-1-4-0	76.33	62.90	PR
PNL 5-8-1-7-21	87.29	76.40	PR
22-2-B-2-1-1	86.46	79.39	PR
92-H 51-4	43.33	32.75	PR
PNL 1-8-5-17-2	92.05	90.56	R
NDR 9830148	90.22	85.32	R
IR 72048-B-R-2-2-2-1-B	83.72	82.58	R
IR 71829-3R-73-1-2-B	94.88	85.72	R
NDRK 5094	88.73	81.78	R
Narendra Usar 3	91.00	90.11	R

Table 2. Pollen viability, spikelet fertility and restoration behaviour of pollen parents with CMS line IR58025A

Pollen parent	Pollen fertility (%)	Spikelet fertility (%)	Restoration behaviour
IR 70023-4B-R-12-3-1-1-B	70.46	57.90	PR
NDRK 5095	73.00	69.09	PR
NDRK 5086	65.28	50.90	PR
NDRK 5013	72.00	67.85	PR
CSRC(S) 14-1-4-0	96.00	66.10	PR
22-2-B-2-1-1	52.10	47.09	PR
92-H 51-4	37.91	25.78	PR
PNL 1-8-5-17-2	94.90	92.33	R
NDRK 5056	99.00	92.84	R
NDR 9830119	93.33	91.79	R
CST 7-1	91.67	91.29	R
21-2-5-B-1-1	95.29	91.42	R
IR 64	92.83	91.50	R
NDR 9830148	93.01	91.14	R
PNL 5-8-1-7-21	96.40	90.99	R
IR 72048-B-R-2-2-2-1-B	93.11	92.74	R
IR 71829-3R-73-1-2-B	95.05	92.79	R
NDRK 5094	94.21	92.01	R
Narendra Usar 3	92.12	90.70	R

R= restorer; PR= partial restorer

The results (Table 1) indicated that the pollen parents taken in this study i.e., IR 61920-3B-22-2-1, IR 70023-4B-R-12-3-1-1-B, NDRK 5095, NDRK 5086, NDRK 5013, CSRC(S) 14-1-4-0, 22-2-B-2-1-1 and 92-H 51-4 were found as partial restorers for both the CMS lines. NDRK 5056, NDR 9830119, CST 7-1, 21-2-5-B-1-1, IR 64 and PNL 5-8-1-7-21 were found as partial restorers to only for NMS 4 A and same testers are found restorers for IR 58025 A. Gautam and Singh (2004) suggested that partial (either restorers nor maintainers) have no utility in the hybrid rice breeding.

Out of 20 pollen parents, PNL 1-8-5-17-2, NDR 9830148, IR 72048-B-R-2-2-2-1-B, IR 71829-3R-73-1-2-B, NDRK 5094 and Narendra Usar 3 were found as complete restorer to both the CMS lines. These genotypes may be helpful for recombination breeding for rapid development of hybrids and broaden the existing genetic base for sodicity tolerance. Similar results are also reported by Chaudhary *et al.* (1982),

Mohanty and Sharma (1983), Tomer and Virmani (1990), Bijral *et al.* (1993), Gautam and Singh (2004), Singh (2005) and Suresh Babu *et al.* (2006) in their studies while using different elite lines.

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