

Development of Component Lines (CMS, Maintainer and Restorer lines) and their Maintenance Using Diversed Cytosources of Rice

Ariful Islam^{1*}, Mian MAK², Rasul G², Bashar K³ and Fatema-Tuj-Johora⁴

¹Department of GPB, EXIM Bank Agricultural University, Bangladesh

²Department of Genetics and Plant Breeding, BSMRAU, Gazipur1706, Bangladesh

³International Potato Research Centre (CIP), Bangladesh

⁴Department of Crop Botany, EXIM Bank Agricultural University, Bangladesh

Abstract

The practice of hybridization has greatly contributed to the increase in crop productivity. A major component that exploits heterosis in crops is the cytoplasmic male sterility (CMS)/nucleus-controlled fertility restoration (*Rf*) system. The development and use of hybrid rice varieties on commercial scale utilizing male sterility and fertility restoration system has proved to be one of the mile stones in the history of rice improvement. Pollen sterility status of 148 exotic rice germplasm was assessed at flowering stage. Sixteen genotypes showed 100% pollen sterility status which was considered as completely male sterile lines (A-line). Sixteen genotypes were also identified as completely fertile due to 80% and above pollen and spikelet fertility. For identification of proper maintainer lines, the identified 16 CMS lines chance crossed with established known maintainer lines viz. IR 58025B, IR 62829B, GAN46B, IR 68888B and BRRI1B. Based on pollen male sterility status of the F_1 s lines it was indicated that 10 out of 16 were maintained by IR 58025B line, 8 CMS lines were maintained by IR 62829B, three CMS were maintained by IR 68888B and one CMS line was maintained by GAN46B and BRRI1B. Restoration potentiality of identified 16 suspected restorer genotypes were assessed through judgement of pollen and spikelet fertility of F_1 s developed through crossing with five standard CMS lines. Based on 80% and above pollen and spikelet fertility of F_1 s, seven suspected restorers were identified as restorer against IR 58025A, two against GAN 46A, five against IR 62829A and two against IR 68888A.

Keywords: Pollen; Spikelet; Fertility; Sterility; Fertility restoration; CMS line; Maintainer line; Synthesized

Introduction

The development and use of hybrid rice varieties on commercial scale utilizing male sterility and fertility restoration system has proved to be one of the mile stones in the history of rice improvement. The hybrid rice technology now in operation, aims at yield increment through higher exploitable heterosis levels [1]. In hybrid rice technology most usually two sterility systems viz., CMS and EGMS are used for commercial seed production. In three line system of hybrid rice variety development system, three lines, A, B and R are required. A line is the cytoplasm-genetic male sterile line where the male sterility is jointly controlled by recessive nuclear gene and sterile cytoplasm. B-line is isogenic line of A-line, only difference in male sterility and fertility. R-line possesses fertility restoration gene [2]. A commercial A-line is characterized by the absence of pollen grains or rudimentary pollens, argonomically superiority, stable sterility, wide regeneration spectrum, abortive anther and highly synchronized [3]. B line is the maintainer line characterized by normal anthers, functional pollens and seed setting on selfing. While normal anthers, functional pollens, abundant pollen producing capacity, strong restoring ability, good combining ability, high out crossing rate, and genetically diverse from CMS line [4,5] are the main characteristics of R-line. It is 30 years since the first commercial release of hybrid rice. Plant cytoplasmic male sterility (CMS), a maternally inherited trait that prevents plants from producing functional pollen, has been identified in many higher plants, including rice, cotton, maize, and sorghum. CMS restorer systems have been widely exploited to produce hybrids that outperform their inbred parents in yield, biomass, or other traits. CMS is usually attributed to an unusual chimeric gene in the mitochondrial genome. In many cases, a nuclear-encoded fertility restorer gene (*Rf*) can restore fertility of the cytoplasmic male-sterile plants. Therefore, the CMS/*Rf* system is an ideal model for dissecting the interaction between mitochondrial and

nuclear genomes. A variety of mechanisms of fertility restoration by the *Rf* genes have been reported for different CMS systems. T-urf13, a mitochondrial gene encoding a 13 kDa protein, has been detected only in maize carrying T male-sterile cytoplasm. The first restorer allele cloned, the maize *Rf2* gene, does not affect the expression of urf13 and encodes aldehyde dehydrogenase (ALDH), which is located in the mitochondrial matrix in a homotetrameric. Hybrid rice has spread such that now it commands about 50% of the total rice area in China but only 7% in Bangladesh [4]. New male sterile cytoplasm sources and inter-subspecies crosses have contributed to the development of super rice breeding. However, sustainable improvements of hybrid rice yield potential, grain quality, and tolerance to biotic and abiotic stresses continue to be a great challenge. Exploitation of new germplasm has always played a critical role in rice breeding, and this will continue [6]. Component lines development is considered as the backbone of any sustainable hybrid rice program. Successful and long lasting hybrid rice program depends on development of diversified component lines utilizing CMS source, local and exotic genetic resources. In this country rice hybrid programs mostly depend on the component lines developed by IRRI and China [7]. There is a shortage of research works in component line development of hybrid rice activities performing by different organizations of Bangladesh [8].

***Corresponding author:** Dr. Ariful Islam M, Department of GPB, EXIM Bank Agricultural University, Bangladesh, Tel: +88-01711872774; E-mail: i.aaarif@yahoo.com

Received February 24, 2015; **Accepted** March 31, 2015; **Published** April 03, 2015

Citation: Islam A, Mian MAK, Rasul G, Bashar K, Johora FT (2015) Development of Component Lines (CMS, Maintainer and Restorer lines) and their Maintenance Using Diversed Cytosources of Rice. J Rice Res 3: 140. doi:10.4172/2375-4338.1000140

Copyright: © 2015 Islam A, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

As a part of that program the present investigation was initiated with the following objectives:

Objectives:

1. Identification of male sterile (CMS) and fertile (B) lines from the exotic rice germplasm.
2. Identification of Restorer (R-lines) based on pollen and spikelet fertility test.

Materials Used

A series of experiments were conducted from Winter (Boro) 2011 to Summer (Aman) 2013 in the experiment field of Department of Genetics and Plant Breeding, Bangabandhu Sheikh Mujibur Rahman

Agricultural University, Gazipur Bangladesh using the following materials (germplasm) (Tables 1 and 2).

Methods of pollen fertility test

Pollen fertility test was done by Potassium Iodide solution (KI). At flowering stage young spikelets were collected early in the morning (7.30-8.50 AM) from the field and kept in the jar for opening the spikelets after about 2 hours. One or two anthers were kept on a glass slide and smashed with KI solution and covered with a cover slip and then observed under a compound Microscope. Records on fertile and sterile pollens from three microscopic focuses were noted. Round well developed stained pollens were considered as viable and non-stained irregular shaped pollens were counted as sterile pollens and then

Genotypes	Genotypes	Genotypes	Genotypes	Genotypes	Genotypes
RG-BU-08-001	RG-BU-08-026	RG-BU-08-051	RG-BU-08-076	RG-BU-08-101	RG-BU-08-126
RG-BU-08-002	RG-BU-08-027	RG-BU-08-052	RG-BU-08-077	RG-BU-08-102	RG-BU-08-127
RG-BU-08-003	RG-BU-08-028	RG-BU-08-053	RG-BU-08-078	RG-BU-08-103	RG-BU-08-128
RG-BU-08-004	RG-BU-08-029	RG-BU-08-054	RG-BU-08-079	RG-BU-08-104	RG-BU-08-129
RG-BU-08-005	RG-BU-08-030	RG-BU-08-055	RG-BU-08-080	RG-BU-08-105	RG-BU-08-130
RG-BU-08-006	RG-BU-08-031	RG-BU-08-056	RG-BU-08-081	RG-BU-08-106	RG-BU-08-131
RG-BU-08-007	RG-BU-08-032	RG-BU-08-057	RG-BU-08-082	RG-BU-08-107	RG-BU-08-132
RG-BU-08-008	RG-BU-08-033	RG-BU-08-058	RG-BU-08-083	RG-BU-08-108	RG-BU-08-133
RG-BU-08-009	RG-BU-08-034	RG-BU-08-059	RG-BU-08-084	RG-BU-08-109	RG-BU-08-134
RG-BU-08-010	RG-BU-08-035	RG-BU-08-060	RG-BU-08-085	RG-BU-08-110	RG-BU-08-135
RG-BU-08-011	RG-BU-08-036	RG-BU-08-061	RG-BU-08-086	RG-BU-08-111	RG-BU-08-136
RG-BU-08-012	RG-BU-08-037	RG-BU-08-062	RG-BU-08-087	RG-BU-08-112	RG-BU-08-137
RG-BU-08-013	RG-BU-08-038	RG-BU-08-063	RG-BU-08-088	RG-BU-08-113	RG-BU-08-138
RG-BU-08-014	RG-BU-08-039	RG-BU-08-064	RG-BU-08-089	RG-BU-08-114	RG-BU-08-139
RG-BU-08-015	RG-BU-08-040	RG-BU-08-065	RG-BU-08-090	RG-BU-08-115	RG-BU-08-140
RG-BU-08-016	RG-BU-08-041	RG-BU-08-066	RG-BU-08-091	RG-BU-08-116	RG-BU-08-141
RG-BU-08-017	RG-BU-08-042	RG-BU-08-067	RG-BU-08-092	RG-BU-08-117	RG-BU-08-142
RG-BU-08-018	RG-BU-08-043	RG-BU-08-068	RG-BU-08-093	RG-BU-08-118	RG-BU-08-143
RG-BU-08-019	RG-BU-08-044	RG-BU-08-069	RG-BU-08-094	RG-BU-08-119	RG-BU-08-144
RG-BU-08-020	RG-BU-08-045	RG-BU-08-070	RG-BU-08-095	RG-BU-08-120	RG-BU-08-145
RG-BU-08-021	RG-BU-08-046	RG-BU-08-071	RG-BU-08-096	RG-BU-08-121	RG-BU-08-146
RG-BU-08-022	RG-BU-08-047	RG-BU-08-072	RG-BU-08-097	RG-BU-08-122	RG-BU-08-147
RG-BU-08-023	RG-BU-08-048	RG-BU-08-073	RG-BU-08-098	RG-BU-08-123	RG-BU-08-148
RG-BU-08-024	RG-BU-08-049	RG-BU-08-074	RG-BU-08-099	RG-BU-08-124	BRR1 Dhan28
RG-BU-08-025	RG-BU-08-050	RG-BU-08-075	RG-BU-08-100	RG-BU-08-125	BRR1 Dhan29

Table 1: 148 Exotic rice genotypes and two checks BRR1 dhan28 and BRR1 dhan29.

Suspected CMS lines	16 suspected R lines	5 known CMS lines	5 known B lines
1. RG BU 08-053 A	1. RG-BU 08-001 R	1. GAN46A	1. GAN46B
2. RG BU 08-058 A	2. RG-BU 08-002 R	2. BRR1A	2. BRR1B
3. RG BU 08-061 A	3. RG-BU 08-005 R	3. IR58025A	3. IR58025B
4. RG BU 08-066 A	4. RG-BU 08-006 R	4. IR62820 A	4. IR62820 B
5. RG BU 08-069 A	5. RG-BU 08-007 R	5. IR68888A	5. IR68888B
6. RG BU 08-084 A	6. RG-BU 08-013 R		
7. RG BU 08-086 A	7. RG-BU 08-016 R		
8. RG BU 08-087 A	8. RG-BU 08-018 R		
9. RG BU 08-107 A	9. RG-BU 08-025 R		
10. RG BU 08-125 A	10. RG-BU 08-034 R		
11. RG BU 08-126 A	11. RG-BU 08-038 R		
12. RG BU 08-129 A	12. RG-BU 08-046 R		
13. RG BU 08-132 A	13. RG-BU 08-057 R		
14. RG BU 08-136 A	14. RG-BU 08-063 R		
15. RG BU 08-137 A	15. RG-BU 08-097 R		
16. RG BU 08-141 A	16. RG-BU 08-105 R		

Table 2: Sixteen suspected CMS lines, 16 suspected restorer lines, 5 established known CMS lines and 5 established known maintainer lines.

converted into percentage. At maturity stage filled spikelets per panicle were counted in each genotype and then converted into percentage.

Data Recorded on

- Pollen fertility
- Spikelet fertility

Identification of A and R lines

The plants having 100% pollens sterility were considered as male sterile (CMS) and the plants having 80% and above pollen fertility and spikelet fertility were initially suspected as restorer line.

Identification of B-lines and maintenance of CMS lines

The identified CMS lines from Chinese germplasm were chance crossed with the established maintainer lines, GAN 46B, BRRI 1B, viz., IR 58025B, IR 62820B and IR 68888B as the corresponding maintainer lines were not known to us. All the 16 CMS lines were crossed with each of the above five maintainer lines. F1 seeds from each cross were harvested separately and grown in next season along with their respective maintainer lines. At flowering stage pollen fertility status of each plant of each cross was recorded. If all the plants of a cross showed 100% pollen sterility then the corresponding male plant was considered as maintainer line.

Performance of restorer lines against established CMS lines

The identified sixteen restorer lines were crossed with five CMS lines viz. GAN46A, BRRI1A, IR58025A, IR62820A and IR68888A. The F1s were grown in the next rice growing season and performance based on pollen and spikelet fertility was analyzed. The F1(s) having above 80% pollen and spikelet fertility indicated that the male parent was an effective restorer.

Results and Discussion

Identification of A and R lines based on Pollen sterility and fertility status

Results of pollen sterility and fertility are shown in Tables 3 and 4.

Among the 148 exotic rice genotypes different levels of pollen sterility and fertility were observed. Such variation in pollen fertility indicated the existence genetic variation in respect of these reproductive traits among the genotypes. Among one hundred and forty eight exotic rice germplasm sixteen genotypes (RG-BU 08-053, RG-BU 08-058, RG-BU 08-061, RG-BU 08-066, RG-BU 08-069, RG-BU 08-084, RG-BU 08-086, RG-BU 08-087, RG-BU 08-107, RG-BU 08-125, RG-BU 08-126, RG-BU 08-129, RG-BU 08-132, RG-BU 08-136, RG-BU 08-137 and RG-BU 08-141) showed 100% pollen sterility status which was considered as completely male sterile lines or A line. The amount is not so less but 10.81% of the total germplasm. Ten genotypes were found sterile having pollen fertility 0-9%. Miyagawa and Nakamura [9] classified 85 rice cultivars based on the regional differences in varietal characteristics and found some elite male fertile lines as well as their counterpart maintainer lines. Chetia et al. [10] evaluated five cytoplasmic male sterile (CMS) rice lines (PMS 2A, PMS 3A, PMS 10A, IR 58025A and IR 62829A) and observed that PMS 2A, PMS 10A and IR 62829A were recorded as complete pollen and spikelet sterility. Sun et al. [11] determined the genetic effects of male sterile cytoplasm on major characters of rice hybrids. Ramesha et al. [12] identified three new and diversified CMS sources and many CMS lines possessing sporophytic type of male sterility with a very high frequency of typically abortive pollen. The new CMS lines were compared with other CMS lines belonging to wild abortive. Besides stable sterility, the new CMS lines had very high panicle exertion rate (92-96%) and good stigma exertion (48-65%) ability. Twenty nine genotypes were recorded partially sterile which is 19.59% of total. But most of the genotypes were categorized in partially fertile based on pollen fertility test. About fifty one genotypes were found partially fertile which 36.46% of total. Such type of genotypes may open the scope of development of restorer lines of hybrid program. And sixteen genotypes (RG-BU 08-001, RG-BU 08-002, RG-BU 08-005, RG-BU 08-006, RG-BU 08-007, RG-BU 08-013, RG-BU 08-016, RG-BU 08-018, RG-BU 08-025, RG-BU 08-034, RG-BU 08-038, RG-BU 08-046, RG-BU 08-057, RG-BU 08-063, RG-BU 08-097 and RG-BU 08-105) were identified as completely fertile as these genotypes had above 80% pollen and spikelet fertility which is 10.81% of the total genotypes. Research results indicated that the

Sl no	Fertility Status	Symbol	Number	Genotypes
1	Completely Sterile	CS	16 (10.81%)	RG-BU 08-053, RG-BU 08-058, RG-BU 08-061, RG-BU 08-066, RG-BU 08-069, RG-BU 08-084, RG-BU 08-086, RG-BU 08-087, RG-BU 08-107, RG-BU 08-125, RG-BU 08-126, RG-BU 08-129, RG-BU 08-132, RG-BU 08-136, RG-BU 08-137 and RG-BU 08-141
2	Sterile	S	10 (6.76%)	RG-BU 08-045, RG-BU 08-049, RG-BU 08-055, RG-BU 08-068, RG-BU 08-072, RG-BU 08-082, RG-BU 08-095, RG-BU 08-099, RG-BU -08-101 and RG-BU 08-122
3	Partially Sterile	PS	29 (19.59%)	RG-BU 08-003, RG-BU 08-015, RG-BU 08-017, RG-BU 08-020, RG-BU 08-030, RG-BU 08-033, RG-BU 08-042, RG-BU 08-043, RG-BU 08-044, RG-BU 08-047, RG-BU 08-056, RG-BU 08-065, RG-BU 08-070, RG-BU 08-077, RG-BU 08-080, RG-BU 08-083, RG-BU 08-092, RG-BU 08-093, RG-BU 08-094, RG-BU 08-106, RG-BU 08-107, RG-BU 08-110, RG-BU -08-115, RG-BU 08-119, RG-BU 08-124, RG-BU 08-134, RG-BU 08-146, RG-BU 08-147 and RG-BU 08-148
4	Partially fertile	PF	51 (34.46%)	RG-BU 08-004, RG-BU 08-009, RG-BU 08-010, RG-BU 08-012, RG-BU 08-014, RG-BU 08-021, RG-BU 08-023, RG-BU 08-026, RG-BU 08-028, RG-BU 08-029, RG-BU 08-031, RG-BU 08-034, RG-BU 08-037, RG-BU 08-039, RG-BU 08-041, RG-BU 08-046, RG-BU 08-048, RG-BU 08-059, RG-BU 08-060, RG-BU 08-062, RG-BU 08-064, RG-BU 08-071, RG-BU 08-073, RG-BU 08-075, RG-BU 08-076, RG-BU 08-078, RG-BU 08-079, RG-BU 08-085, RG-BU 08-088, RG-BU 08-091, RG-BU 08-096, RG-BU 08-098, RG-BU 08-101, RG-BU 08-102, RG-BU 08-104, RG-BU 08-105, RG-BU 08-109, RG-BU 08-112, RG-BU 08-113, RG-BU 08-114, RG-BU 08-118, RG-BU 08-121, RG-BU 08-123, RG-BU 08-128, RG-BU 08-131, RG-BU 08-133, RG-BU 08-135, RG-BU 08-140, RG-BU 08-141, RG-BU 08-143 and RG-BU 08-145,
5	Fertile	F	26 (17.57%)	RG-BU 08-008, RG-BU 08-011, RG-BU 08-019, RG-BU 08-024, RG-BU 08-033, RG-BU 08-035, RG-BU 08-040, RG-BU 08-042, RG-BU 08-043, RG-BU 08-050, RG-BU 08-051, RG-BU 08-052, RG-BU 08-054, RG-BU 08-056, RG-BU 08-074, RG-BU 08-085, RG-BU 08-088, RG-BU 08-090, RG-BU 08-096, RG-BU 08-100, RG-BU 08-103, RG-BU 08-117, RG-BU 08-128, RG-BU 08-131, RG-BU -08-139 and RG-BU 08-146
6	Highly/fully Fertile	FF	16 (10.81%)	RG-BU 08-001, RG-BU 08-002, RG-BU 08-005, RG-BU 08-006, RG-BU 08-007, RG-BU 08-013, RG-BU 08-016, RG-BU 08-018, RG-BU 08-025, RG-BU 08-034, RG-BU 08-038, RG-BU 08-046, RG-BU 08-057, RG-BU 08-063, RG-BU 08-097 and RG-BU 08-105

Table 3: Classification of 148 (Chinese) exotic rice germplasm based on pollen fertility status.

Pollen fertility status	Fertility Percent
1. FS: Fully Sterile/Completely sterile	0%
2. HS: Sterile	1-9%
3. PS: Partially Sterile	10-29%
4. HF: Partially Fertile	30-69%
5. F: Fertile	70-79%
6. FF: Fully Fertile	80 % and above

Table 4: Classification of 148 (Chinese) exotic rice germplasm based on pollen fertility status.

frequency of restorer lines in Chinese rice germplasm was found very high as compared to local rice genotypes. Thus the above mentioned genotypes having restorer genes may be utilized as a good reservoir of restorer genes for development of efficient restorer lines. Abeysekera et al. [13] studied 53 cytoplasmic male sterile (CMS) lines of rice and observed that pollen fertility and spikelet fertility had significant effects on the out crossing rate (Tables 3 and 4).

Restorability of suspected restorer lines

Sixteen suspected restorer lines were used to test their restoration ability with four established CMS lines, IR 58025A, GAN 46A, IR 62820A and IR 68888A. Results of restoration ability based on pollen and spikelet fertility of F₁s between CMS and restorer lines are shown in Tables 5 and 6. Out of sixteen F₁ ten crosses (IR 58025A×RG-BU 08-001R, IR 58025A×RG-BU 08-006R, IR 58025A×RG-BU 08-013R, IR 58025A×RG-BU 08-016R, IR 58025A×RG-BU 08-018R, IR 58025A×RG-BU 08-025R, IR 58025A×RG-BU 08-034R, IR 58025A×RG-BU 08-046R, IR 58025A×RG-BU 08-063R and IR 58025A×RG-BU 08-0105R) showed above 80% pollen fertility but seven crosses showed 80% above both pollen and spikelet's fertility (IR 58025A×RG-BU 08-001R, IR 58025A×RG-BU 08-006R, IR 58025A×RG-BU 08-018R, IR 58025A×RG-BU 08-034R, IR 58025A×RG-BU 08-046R, IR 58025A×RG-BU 08-063R and IR 58025A×RG-BU 08-0105R) which indicated that seven suspected restorers had desirable restoration

ability with the above CMS line. When these sixteen suspected restorers were crossed with GAN 46A six F₁s (GAN46A×RG-BU 08-007R, GAN46A×RG-BU08-025R, GAN46A×RG-BU 08-034R, GAN46A×RG-BU 08-038R, GAN46A×RG-BU 08-097R and GAN46A×RG-BU 08-105R) showed above 80% pollen fertility but only two crosses (GAN46A×RG-BU 08-007R and GAN46A×RG-BU08-025R) showed 80% above both pollen and spikelet's fertility. Eight crosses of restorer lines with IR 62820A showed above 80% pollen fertility (IR 62820A×RG-BU 08-001R, IR 62820A×RG-BU 08-005R, IR 62820A×RG-BU 08-006R, IR 62820A×RG-BU 08-007R, IR 62820A×RG-BU 08-013R, IR 62820A×RG-BU 08-025R, IR 62820A×RG-BU 08-038R and IR 62820A×RG-BU 08-057R) where as five F₁s were found having 80% and above both pollen and spikelet's fertility i.e., IR 62820A×RG-BU 08-005R, IR 62820A×RG-BU 08-013R, IR 62820A×RG-BU 08-025R, IR 62820A×RG-BU 08-038R and IR 62820A×RG-BU 08-057R. Five crosses of restorer lines with IR 68888A showed above 80% pollen fertility (IR 62820A×RG-BU 08-001R, IR 62820A×RG-BU 08-002R, IR 62820A×RG-BU 08-007R, IR 62820A×RG-BU 08-097R and IR 62820A×RG-BU 08-105R) where as only two F₁s were found having 80% and above both pollen and spikelet's fertility i.e., IR 62820A×RG-BU 08-002R and IR 62820A×RG-BU 08-007R. Li et al. [14] identified R899 as an early-maturing restorer line. Its characteristics include desirable agronomic traits, strong restoring ability, strong resistance to rice blast, fine grain quality and high hybrid seed yield. The hybrid combinations displayed high and stable grain yield, fine grain quality, suitable growth period and wide adaptability. Five out of sixteen F₁ crosses with IR 62820A showed above 80% pollen and spikelet fertility. Such results indicated that only five lines have desirable restoration ability in hybrids with IR 62820A. In case of IR 68888A only two restorers were found effective [15-17]. Further study on the performance of F₁ hybrids obtained from crosses between IR 62820A and IR 68888A and the selected five and two restorer lines may done for identification of desirable rice hybrid.

Test crosses	IR 58025B		BRR1 1B		GAN 46B		IR 62820B		IR 68888B	
	Pollen Fertility (PF)	Spikelet's fertility (SF)	Pollen Fertility (PF)	Spikelet's fertility (SF)	Pollen Fertility (PF)	Spikelet's fertility (SF)	Pollen Fertility (PF)	Spikelet's fertility (SF)	Pollen Fertility (PF)	Spikelet's fertility (SF)
RG BU 08-053 A	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	3.00 ± 0.32	12.40 ± 0.34	13.00 ± 0.32	29.12 ± 0.37
RG BU 08-058 A	7.00 ± 0.14	15.68 ± 0.17	6.00 ± 0.27	12.44 ± 0.27	6.00 ± 0.27	12.44 ± 0.32	9.00 ± 0.27	20.16 ± 0.32	11.00 ± 0.32	24.64 ± 0.32
RG BU 08-061 A	8.00 ± 0.13	17.92 ± 0.18	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	8.00 ± 0.27	17.92 ± 0.32	13.00 ± 0.32	29.12 ± 0.31
RG BU 08-066 A	11.00 ± 0.15	24.64 ± 1.19	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
RG BU 08-069 A	12.00 ± 0.19	26.88 ± 4.15	6.00 ± 0.24	13.44 ± 0.27	6.00 ± 0.27	13.44 ± 0.27	9.00 ± 0.27	20.16 ± 0.32	5.00 ± 0.27	11.20 ± 0.32
RG BU 08-084 A	0.00 ± 0.00	0.00 ± 0.00	8.00 ± 0.27	17.92 ± 0.32	8.00 ± 0.23	17.92 ± 0.32	0.00 ± 0.00	0.00 ± 0.00	6.00 ± 0.27	13.44 ± 0.32
RG BU 08-086 A	0.00 ± 0.00	0.00 ± 0.00	9.00 ± 0.26	20.16 ± 0.32	9.00 ± 0.22	20.16 ± 0.32	23.00 ± 0.19	21.52 ± 0.32	9.00 ± 0.27	20.16 ± 0.32
RG BU 08-087 A	0.00 ± 0.00	0.00 ± 0.00	12.00 ± 0.32	26.88 ± 2.12	12.00 ± 0.32	26.88 ± 0.32	0.00 ± 0.00	0.00 ± 0.00	9.00 ± 0.27	20.16 ± 0.32
RG BU 08-107 A	0.00 ± 0.00	0.00 ± 0.00	13.00 ± 0.32	29.12 ± 2.12	13.00 ± 0.31	29.12 ± 2.19	0.00 ± 0.00	0.00 ± 0.00	8.00 ± 0.27	17.92 ± 0.32
RG BU 08-125 A	0.00 ± 0.00	0.00 ± 0.00	8.00 ± 0.24	17.92 ± 0.32	8.00 ± 0.27	17.92 ± 0.32	9.00 ± 0.27	20.16 ± 0.32	12.00 ± 0.32	26.88 ± 0.32
RG BU 08-126 A	9.00 ± 0.15	20.16 ± 2.19	9.00 ± 0.27	20.16 ± 0.32	9.00 ± 0.21	20.16 ± 0.32	8.00 ± 0.27	17.92 ± 0.32	1.22 ± 0.27	3.54 ± 0.27
RG BU 08-129 A	0.00 ± 0.00	0.00 ± 0.00	7.00 ± 0.22	15.68 ± 0.27	7.00 ± 0.27	15.68 ± 0.32	0.00 ± 0.00	0.00 ± 0.00	3.00 ± 0.27	6.72 ± 0.27
RG BU 08-132 A	8.00 ± 0.16	17.92 ± 0.87	13.00 ± 0.32	29.12 ± 2.10	13.00 ± 0.32	29.12 ± 2.19	7.00 ± 0.27	15.68 ± 0.32	14.00 ± 0.32	31.36 ± 0.39
RG BU 08-136 A	0.00 ± 0.00	0.00 ± 0.00	14.00 ± 0.32	31.36 ± 2.10	14.00 ± 0.32	35.72 ± 2.19	0.00 ± 0.00	0.00 ± 0.00	11.00 ± 0.32	24.64 ± 0.32
RG BU 08-137 A	0.00 ± 0.00	0.00 ± 0.00	18.00 ± 0.38	20.32 ± 2.12	18.00 ± 0.32	20.32 ± 0.32	0.00 ± 0.00	0.00 ± 0.00	11.00 ± 0.32	24.64 ± 0.32
RG BU 08-141 A	0.00 ± 0.00	0.00 ± 0.00	22.00 ± 2.11	29.28 ± 2.19	22.00 ± 1.19	29.28 ± 2.19	0.00 ± 0.00	0.00 ± 0.00	10.00 ± 0.32	22.40 ± 0.32
Minimum	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Maximum	12.00	26.88	23.00	31.36	23.00	35.72	23.0	22.4	13.00	31.36
Average	7.06	14.31	7.06	14.31	7.06	14.31	8.37	10.52	7.06	14.31
SD	6.05	10.90	6.05	10.90	6.05	10.90	7.87	5.24	6.05	10.90
SE	0.67	1.21	0.67	1.21	0.67	1.22	1.25	1.43	0.67	1.19
R ²	-0.06	-0.04	-0.06	-0.05	-0.06	-0.04	-0.04	-0.03	-0.07	-0.04

Table 5: Maintenance of identified CMS lines using IR 58025B, BRR1 1B, GAN 46B, IR 62820B and IR 68888B.

Test crosses	IR 58025A		BRRI 1A		GAN 46A		IR 62820A		IR 68888A	
	Pollen Fertility (PF)	Spikelet's fertility (SF)	Pollen Fertility (PF)	Spikelet's fertility (SF)	Pollen Fertility (PF)	Spikelet's fertility (SF)	Pollen Fertility (PF)	Spikelet's fertility (SF)	Pollen Fertility (PF)	Spikelet's fertility (SF)
RG-BU 08-001 R	100.00 ± 0.00	87.35 ± 4.10	12.12 ± 1.19	34.77 ± 4.19	71.16 ± 4.36	56.00 ± 4.19	87.45 ± 4.19	56.99 ± 4.19	91.12 ± 4.36	43.12 ± 4.19
RG-BU 08-002 R	76.44 ± 4.19	54.45 ± 4.11	3.32 ± 2.19	13.13 ± 3.19	7.12 ± 0.32	12.12 ± 4.19	34.34 ± 4.10	43.12 ± 4.10	90.00 ± 4.36	89.01 ± 4.36
RG-BU 08-005 R	64.87 ± 4.19	47.77 ± 4.19	100.00 ± 0.00	85.11 ± 4.36	63.23 ± 4.36	45.45 ± 4.19	100.00 ± 0.00	92.65 ± 4.36	12.12 ± 1.19	34.77 ± 4.19
RG-BU 08-006 R	100.00 ± 0.00	92.53 ± 4.36	12.00 ± 4.33	84.44 ± 4.36	9.23 ± 0.32	12.34 ± 4.19	95.34 ± 4.36	34.77 ± 4.19	3.32 ± 2.19	13.13 ± 3.19
RG-BU 08-007 R	76.23 ± 4.36	34.34 ± 4.19	78.78 ± 4.36	68.94 ± 4.36	100.00 ± 0.00	87.57 ± 4.36	91.12 ± 4.36	31.77 ± 4.18	100.00 ± 0.00	85.11 ± 4.36
RG-BU 08-013 R	98.87 ± 4.36	56.44 ± 4.19	78.90 ± 4.36	81.76 ± 4.19	34.35 ± 4.19	46.49 ± 4.19	100.00 ± 0.00	83.88 ± 4.36	12.00 ± 4.33	84.44 ± 4.36
RG-BU 08-016 R	91.23 ± 4.36	56.56 ± 4.19	78.00 ± 4.36	40.01 ± 4.17	34.00 ± 4.19	48.48 ± 4.19	45.90 ± 4.19	56.56 ± 4.19	78.78 ± 4.36	68.94 ± 4.36
RG-BU 08-018 R	100.00 ± 0.00	86.34 ± 4.36	71.00 ± 4.19	82.99 ± 4.36	56.34 ± 4.19	82.25 ± 4.36	68.68 ± 4.19	71.17 ± 4.19	78.90 ± 4.36	81.76 ± 4.19
RG-BU 08-025 R	92.22 ± 4.36	67.67 ± 4.19	61.00 ± 4.19	53.77 ± 4.19	98.34 ± 4.36	82.76 ± 4.36	100.00 ± 0.00	83.99 ± 4.36	78.00 ± 4.36	40.01 ± 4.17
RG-BU 08-034 R	100.00 ± 0.00	87.09 ± 4.36	63.00 ± 4.19	81.09 ± 4.36	90.24 ± 4.36	56.00 ± 4.12	67.99 ± 4.19	42.24 ± 4.17	71.00 ± 4.19	82.99 ± 4.36
RG-BU 08-038 R	69.23 ± 4.19	34.88 ± 4.19	78.00 ± 4.19	74.26 ± 4.36	90.33 ± 4.36	32.12 ± 4.12	100.00 ± 0.00	85.22 ± 4.36	61.00 ± 4.19	53.77 ± 4.19
RG-BU 08-046 R	100.00 ± 0.00	91.78 ± 4.36	67.66 ± 4.19	46.25 ± 4.10	23.23 ± 4.10	45.45 ± 4.12	71.11 ± 4.19	56.00 ± 4.10	63.00 ± 4.19	81.09 ± 4.36
RG-BU 08-057 R	12.12 ± 0.32	23.23 ± 4.19	90.99 ± 4.36	54.76 ± 4.19	34.34 ± 4.10	42.34 ± 4.12	100.00 ± 0.00	83.32 ± 4.36	78.00 ± 4.19	74.26 ± 4.36
RG-BU 08-063 R	100.00 ± 0.00	92.65 ± 4.36	98.00 ± 4.36	40.89 ± 4.10	76.23 ± 4.36	87.57 ± 4.36	34.34 ± 4.19	76.49 ± 4.36	67.66 ± 4.19	46.25 ± 4.10
RG-BU 08-097 R	10.23 ± 0.32	23.00 ± 4.19	91.12 ± 4.36	43.12 ± 4.19	100.00 ± 0.00	76.49 ± 4.36	34.98 ± 4.19	48.48 ± 4.13	90.99 ± 4.36	54.76 ± 4.19
RG-BU 08-105 R	100.00 ± 0.00	82.76 ± 4.36	90.00 ± 4.36	89.01 ± 4.36	98.48 ± 4.36	48.48 ± 4.19	45.22 ± 4.19	56.99 ± 4.19	98.00 ± 4.36	40.89 ± 4.10
Minimum	2.672	12.123	2.673	12.127	7.172	12.123	2.672	12.123	3.324	13.127
Maximum	100.000	92.654	100.000	92.656	100.000	92.654	100.000	92.654	100.000	89.065
Average	67.847	60.803	67.841	60.806	67.847	60.803	67.847	60.803	65.846	60.806
SD	32.384	22.564	32.387	22.567	32.384	22.564	32.384	22.564	32.387	22.567
SE	3.604	2.513	3.608	2.516	3.604	2.513	3.604	2.513	3.608	2.516
R ²	0.752	0.672	0.752	0.672	0.752	0.672	0.729	0.672	0.695	0.672

Table 6: Fertility Restoration of identified R-lines against IR 58025A, BRRI 1A, GAN 46A, IR 62820A and IR 68888A.

References

- Islam MA, Mian MAK, Rasul G, Johora, Sarker UK (2010) Interaction effect between genotypes, row ratio and fertilizer dose on hybrid seed production of rice (*Oryza sativa* L.). Bangladesh Society of Agronomy 4: 134-141.
- Islam MA, Mian MA, Rasul G, Khaliq QA, Bashar MK (2014) Estimation of specific combining ability (SCA) effects and per-se performances in some reproductive traits in rice (*Oryza sativa* L.). Bangladesh Journal of Genetics and Plant Breeding 33: 12-16.
- Yuan LP (1998) Genetic relationship of stigma exterior between maintainer lines and sterile lines for Dian type japonica hybrid rice. Journal-of-Yunnan-Agricultural-University 20: 459-461, 477.
- Islam MA, Sarker UK, Mian MAK, Ahmed JU (2009) Genotype Seedling age Interaction for hybrid seed yield of rice (*Oryza sativa* L.). Bangladesh Journal of Genetics and Plant Breeding 24: 23-26.
- Mian MAK (2010) Hybrid rice breeding. Breeding for self-pollinated crops, Heterosis Breeding. BSMRAU, Summer.
- Islam MA, Mian MAK, Rasul G, Khaliq QA, Bashar MK (2014) Estimation of general combining ability (GCA) effects and per-se performances in some reproductive traits in rice (*Oryza sativa* L.). Bangladesh Journal of Genetics and Plant Breeding 32: 18-13.
- Islam MA, Johora FT, Sarker UK, Mian MAK (2012) Adaptation of Chinese CMS lines interaction with seedling age and row ratio on hybrid seed production of Rice (*Oryza sativa* L.). Bangladesh Journal of Agronomy 25: 178-183.
- Islam MA, Johora FT, Hasan MR, Islam MR, Mahmud MNH (2012) Optimizing sowing date of morpho-physiological traits in direct seeding and transplanting rice (*Oryza sativa* L.). Echo-Friendly Agriculture Journal 5: 162-167.
- Miyagawa S, Nakamura S (1994) Regional differences in varietal characteristics of scented rice. Japan J. crop Sci 53: 494-502.
- Chitra S, Awan E, Ijaz M, Manzoor Z (2000) Heterosis and combining ability analysis in Basmati rice hybrids. J Animal and Pl Sci 16: 56-59.
- Sun A, Kreetapiron S, Varayanond W, Tungtrakul P, Somboonpong S, Rattapat S (2006) Combining ability analysis for grain yield and its components in rice Karnataka J Agric Sci 16: 223-227.
- Ramesha MS, Ahmed MI, Viraktamath BC, Vijayakumar CHM, Singh S (2008) New cytoplasmic male sterile (CMS) lines with diversified CMS sources and better out crossing traits in rice. Intl. Rice Res. Notes.23: 5.
- Abeysekera LPR, Saini G, Sharma AK (2003) Studies on morphological traits of rice (*Oryza sativa* L.). New-Agriculturist 7: 79-83
- Li ZK, Luo LJ, Mei HW, Wang DL, Shu QY, Tabien R et al., (2006) Overdominant epistatic loci are the primary genetic basis of inbreeding depression and heterosis in rice. I. Biomass and grain yield. Genetics 158 : 1737-1753.
- Hien NL, Sarhadi WA, Oikawa Y, Hirata Yi (2007) Genetic diversity of morphological responses and the relationships among Asian rice (*Oryza sativa* L.) cultivars. Tropics 16: 343-355.
- Islam MA, Mian MAK, Rasul G, Khaliq QA, Bashar MK (2013) Estimation of Specific Combining Ability (GCA) Effects and Per-se performances in Some Yield Related Traits of Rice (*Oryza sativa* L.). Echo-Friendly Agriculture Journal 7: 143-145.
- Rita KT, Sarawgi A (2008) Differential fertility restoration of restorer genes to WA-cytoplasmic male sterility system in rice (*Oryza sativa* L.). Indian-Journal-of-Genetics-and-Plant-Breeding; 65: 207-208.