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Short Communication

Development and characterization of fertility restorers in cytoplasmic genetic male sterile lines of cotton (*Gossypium hirsutum* L.) derived from *Gossypium harknessii*

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Cytoplasmic-nuclear male sterility (CGMS) is a maternally inherited trait and in conjunction with nuclear genome suppresses the production of viable pollen grains without affecting the female fertility. Male sterile plants are able to set seeds as long as viable pollens are provided. The presence of certain nuclear genes Rf (restoring fertility) can effectively suppress the male-sterile cytoplasm and restore pollen fertility. However, the success in development of cotton hybrid largely depends on availability of the effective restorer and precise basic knowledge on the genetics of fertility restoration of such lines. The first F_1 line of commercial cotton was introduced by crossing an upland cotton (*G. hirsutum*) as a male parent to a wild species *G. harknessii* [1]. This system is involving cytoplasmic male sterility (A), maintainer (B) and restorer (R) lines, has been widely used to develop hybrid rice varieties [2, 3]. Reduction in cost of hybrid seed production is possible by using male sterility. The best-known sterile cytoplasmic source available for heterosis breeding in cotton is from *G. harknessii* developed by Meyer [4, 5]. The CMS system could not be exploited commercially because of complex nature of fertility restoration and a suspected penalty on seed cotton yield and yield contributing traits as reported by several workers [6-9]. The reduction in yield and its component traits may be due to the less compatibility of alien cytoplasm with the nuclear genes of *G. hirsutum.* But by the course of evolution this incompatibility can be overcome. By keeping this in view, breeding programme to develop alloplasmic restorer lines using pedigree method was initiated.

The CMS system developed by Meyer [10] has been used to develop cytoplasmic male sterile lines. Four cytoplasmic genic male sterile alloplasmic lines (CGMS) lines *viz.*, CMS SPC 1, CMS SPC 5, CMS SPC 9, CMS SPC 11, were developed at CICR Regional Station, Sirsa using IH 76 carrying *G. harknessii* cytoplasm by back cross breeding. These CMS lines were crossed with restorer euplasmic lines Cotton Institute Restorers (CIR-7, CIR-9, CIR-20, CIR-26 and CIR-69). The resulting F_1 were evaluated for yield and quality traits, It was found that *G. harknessii* cytoplasm source suppresses the yield, ginning, fibre fineness etc. To develop the new restorer lines, the outstanding F_1s were selfed to produce F_2. The outstanding fertile plants were selected from the F_2 and subsequent generations. Selected plants were covered to obtain selfed seeds in each generation. The same procedure was followed up to F_7 generations. In this way 25 fertility restorer lines namely, CIR 97 $P_{1\text{-}1}$, CIR 97 $P_{1\text{-}3}$, CIR 97 $P_{1\text{-}4}$, CIR 97 P_2, CIR 97 $P_{1\text{-}4}$, CIR 97 P_2, CIR 97 $P_{3\text{-}1}$, CIR 97 $P_{3}{}'{}_2$, CIR 97 $P_{3}{}'{}_3$, CIR 97 $P_{3\text{-}3}$, CIR 97 $P_{3}{}'{}_5$, CIR 119 $P_{1\text{-}1}$, CIR 119 $P_{1\text{-}2}$, CIR 119 $P_{1\text{-}3}$, CIR 119 $P_{2\text{-}1}$, CIR 126 $P_{1\text{-}2}$, CIR 126 $P_{2}{}'{}_2$, CIR 126 P_3, CIR 526 P_1, CIR 526 P_2, CIR 526 P_3, CIR 920 $P_{1\text{-}2}$, CIR 920 $P_{1\text{-}3}$, CIR 926 $P_{2\text{-}1}$ and CIR 926 $P_{2}{}'{}_3$, were developed which have alien cytoplasm of *G. harknessii* and have the ability to restore fertility. These fertility restorers were evaluated and characterized during *kharif* season 2006-2007. On the basis of pollen dehiscence plants were classified as male fertile or male sterile. After stabilization of restorer lines, they were tested in three rows of 6 m row length for two years. The selections, which were found stable for fertility

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restoration, were characterized for agro-morphological characters. The observations on agro-morphological characters on five randomly selected plants of each restorer lines for two consequently years were made and averages are presented.

The twenty five restorer lines developed by using pedigree breeding approach were characterized on the bases of following morphological characters (Table 1). *Viz*., leaf shape, leaf surface, stem colour, flower colour, anther colour, and seed fuzz colour. Except few most of the restorer lines possessed one or more peculiar characteristics which differentiate them from each other. So on the bases of morphological traits, they can be identified easily and these traits may be used in screening and identification of the particular restorer line, if it is used in hybrid seed production. Restorer lines CIR 97 P_2 and CIR 526 P_3 possess okra type leaves, which make them distinguishable from other restorer lines. Similarly three restorer lines CIR 97 P_2, CIR 97 P_{1-4} and CIR 526 P_3 possesses glabrous leaf surface which can be serve as phenotypic marker in identification of these lines at early seedling stage. Likewise restorer lines CIR 97 P_{1-4}, CIR 97 P_{1-3}, CIR 97 P_{3-3} and CIR 526 P_2 have red stem and leaf with bicolour flower colour which makes them identifiable from other restorer lines even at seedling stage. The above discussed traits can be used as morphological markers to identify restorer lines at the seedling stage of plant development. Similarly flower, anther and fuzz colour can also serve as morphological markers at post flowering stage of plant development and for these traits the restorer lines differentiate distinguishably.

Table 1. Characteristics of newly developed restorer lines

Restorer lines	Characters					
	Leaf shape	Leaf surface	Flower color	Anther color	Stem color	Seed fuzz colour
1. CIR 97 P_{1-1}	N	H	CF	CC	GP	White
2. CIR 97 P_{1-3}	N	H	CF	CC	GP	White
3. CIR 97 P_{1-2}	N	H	CF	CC	GP	White
4. CIR 97 P_{1-4}	N	GL	CF	CC	RP	Grey
5. CIR 97 P_2	OL	GL	CF	CC	GP	Grey
6. CIR 97 P_{3-1}	N	H	YR	CC	RP	Grey
7. CIR 97 P_{3-2}	N	H	YF	YC	GP	Grey
8. CIR 97 P_{3-3}	N	H	YR	CC	RP	Grey
9. CIR 97 P_{3-4}	N	H	CF	CC	GP	Grey
10. CIR 97 P_{3-5}	N	H	CF	CC	GP	Grey
11. CIR 97 P_{4-4}	N	H	CF	CC	GP	White
12. CIR 119 P_{1-1}	N	H	YF	YC	GP	White
13. CIR 119 P_{1-2}	N	H	YF	YC	GP	Grey
14. CIR 119 P_{1-3}	N	H	CF	YC	GP	Grey
15. CIR 119 P_{2-1}	N	H	CF	YC	GP	Grey
16. CIR 126 P_{1-2}	N	H	CF	CC	GP	White
17. CIR 126 P_{2-2}	N	H	CF	LY	GP	White
18. CIR 126 P_3	N	H	CF	CC	GP	White
19. CIR 526 P_1	N	H	CF	CC	GP	White
20. CIR 526 P_2	N	H	CR	LY	RP	White
21. CIR 526 P_3	OL	GL	YF	LY	GP	Grey
22. CIR 920 P_{1-2}	N	H	CF	CC	GP	White
23. CIR 920 P_{1-3}	N	H	CF	CC	GP	White
24. CIR 926 P_{2-1}	N	H	CF	CC	GP	White
25. CIR 926 P_{2-3}	N	H	CF	CC	GP	White

CLCV = cotton leaf curl virus; N = normal leaf; OL okra leaf; H = hairiness; GL = glabrous; CF = cream flower; YF = yellow flower; YR = yellow red; CR = cream red; LY light yellow,. RP = Red plant; GP = Green plant; RC = Red colour; YC = yellow colour; CC = cream colour

The restorer lines were evaluated for yield and contributing traits *viz*., plant height, number of monopods, number of sympods/plant, number of bolls/plant, boll weight/plant, GOT% and seed index.

Twenty five phenotypically diverse restorers were characterized for agro morphological traits and designated as Cotton Institute Restorers (CIR) as mentioned in Table 2. Plant height ranged from 83 cm (CIR 920 P_{1-2}) to 160 cm (CIR 119 P_{1-3}). Number of monopods and sympods also ranged from 0.0 (CIR 920 P_{1-3}) to 5.0 (CIR 97 P_2) and 9.6 (CIR 97 P_3) to 19.6 (CIR 97 P_4) respectively. Similarly number of bolls/plant ranged from 24 (CIR 920 P_{2-2}) to 59.4 (CIR 97 P_{3-5}). Restorer line CIR 126 P_{2-2}, depicted heaviest boll weight (4.0 g), while the same was lightest in line number CIR 97 P_{3-3}. GOT% ranged from 28.0 (CIR 119 P_{2-1}) to 36.4 (CIR 119 P_{1-2}) and the seed index range was from 5.5 g (CIR 97 P_{3-3}) to 10.0 g (CIR 126 P_{2-2}).

Weaver and Weaver [11] observed two deleterious traits in population carrying gene for fertility restoration. One of these was cracked root, which causes the roots to be severely cracked and underdeveloped. Another deleterious factor was characterized by dwarfed and distorted terminal leaves. Therefore, this gives the clue to intermate the different newly identified restorer lines, so that level of fertility restoration can be enhanced due to recombination of different genes [9, 11]. The restorer lines, which have shown stable restorability and are

Table 2. Evaluation of newly developed restorer lines based on yield and associated traits

S.No.	CMS lines	Plant height (cm)	No. of monopods	No. of sympods	Bolls/ plant	Boll weight (g)	GOT (%)	Seed index (g)
1	CIR 97 P_{1-1}	145	2.0	19.0	53.40	3.40	28.8	7.5
2	CIR 97 P_{1-3}	122	2.6	9.0	45.70	2.50	32.4	8.4
3	CIR 97 P_{1-2}	145	1.0	18.0	43.00	2.80	33.4	7.9
4	CIR 97 P_{1-4}	143	1.7	19.6	43.20	3.60	35.2	7.5
5	CIR 97 P_{2}	159	5.0	16.6	48.00	3.05	32.6	6.8
6	CIR 97 P_{3-1}	147	1.0	12.3	31.70	2.90	33.6	6.7
7	CIR 97 P_{3-2}	159	3.3	16.0	38.00	2.70	31.6	6.7
8	CIR 97 P_{3-3}	130	2.0	13.0	45.20	2.45	33.6	5.5
9	CIR 97 P_{3-4}	129	2.0	15.3	43.00	2.55	32.4	5.9
10	CIR 97 P_{35}	135	2.3	13.3	59.40	2.75	33.0	7.2
11	CIR 97 P_{44}	127	1.6	14.3	48.80	2.70	33.0	7.7
12	CIR 119 P_{1-2}	140	3.0	14.0	30.00	3.04	33.8	7.9
13	CIR 119 P_{1-2}	146	1.5	14.6	43.33	3.75	36.4	7.4
14	CIR 119 P_{1-3}	160	3.3	17.3	46.40	2.70	33.4	6.8
15	CIR 119 P_{2-1}	153	3.3	16.0	54.00	2.70	28.0	8.0
16	CIR 126 P_{1-2}	135	2.0	16.3	30.66	3.00	29.0	8.5
17	CIR 126 P_{2-2}	130	3.2	13.0	32.33	4.00	30.6	10.0
18	CIR 126 P_{3}	103	3.0	12.3	41.00	3.20	29.2	9.2
19	CIR 526 P_{1}	118	2.2	10.3	27.40	3.50	35.6	9.6
20	CIR 526 P_{2}	139	4.0	12.6	29.60	3.50	30.8	8.0
21	CIR 526 P_{3}	124	3.6	9.6	43.40	3.90	32.6	8.5
22	CIR 920 P_{1-2}	83	1.0	11.0	24.00	3.30	32.0	8.8
23	CIR 920 P_{1-3}	118	0.0	17.6	32.00	3.40	33.6	7.0
24	CIR 926 P_{2-1}	117	1.6	12.0	33.40	3.00	32.4	8.0
25	CIR 926 P_{2-3}	113	2.0	16.0	45.70	2.80	30.2	7.7

agronomically desirable types, can be utilized for development of cotton hybrids based on cytoplasmic genetic male sterility system. However no such deleterious effects were observed among the restorer lines developed in the present study. The use of these restorer lines should permit the commercial production of completely fertile FI cotton hybrids.

References

1. **Meyer V. G.** 1973. Fertility restorer genes for cytoplasmic male-sterility from *Gossypium harknessii.* Beltwide Cotton Prod. Res. Conf. Proc., pp 65.
2. **Lin S. C. and Yuan L. P.** 1980. Hybrid rice breeding in China. Pp. 35-51, *Oryza sativa* L. II. The inheritance of male sterility. Japanese J. Genetics., **44:** 149-156.
3. **Virmani S.S. and Banghui W.** 1988. Development of CMS lines in hybrid rice breeding. *In*: Hybrid Rice, pp. 103-114, IRRI, P.O. Box 933, Manila, Philippines.
4. **Meyer V. G.** 1965. Cytoplasm effects on anther numbers in interspecific hybrids. J. Hered., **56**: 292-294.
5. **Meyer V.** G.I975. Male sterility from *Gossypium harknessii.* J. Hered., **66**: 23-27.
6. **Weaver Jr. J. B.** 1986. Performance of open pollinated cultivars and CMS upland x upland restorer lines. Proc. Beltwide Cotton Prod. Res. Con., 98-99.
7. **Wei Z. G., Li Z.Y., Hua J. P. and Yi X. D.** 1995. Studies of effects of the male sterile cytoplasm of G. *harknessii.* Acta Gossypii Sinica, **11**: 268-274.
8. **Zhu X. F., Wang X. D., Sun J., Zhang T. Z. and Pan J. J.** 1998. Assessment of cytoplasmic effects of male sterile lines in upland cotton. Pl. Breed., **117**: 549-552.
9. **Tuteja O. P., Kumar Sunil and Singh Mahendar.** 2004 a. Identification of new fertility restorers based on cytoplasmic male sterility system in cotton (*G. hirsutum*). *In:* Strategies for Sustainable Cotton Production: A Global Vision 1. Crop Improvement. International Symposium, 23-25 November, 2004, University of Agricultural Sciences, Dharwad: 107-109.
10. **Meyer V. G.** 1971. Cytoplasmic effect on anther number in interspecific hybrids of cotton. J. Hered., **62**: 77-78.
11. **Weaver D. B. and Weaver J.B. Jr.** 1977. Inheritance of pollen fertility restoration in cytoplasmic male sterile upland cotton. Crop Sci., **17:** 497-499.