



## Identification of fertility restorers for *Gossypium harknessii* based cytoplasmic male sterility system in cotton (*Gossypium hirsutum* L.)

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Cytoplasmic male sterility (CMS) is a maternally inherited trait conferring the inability to produce functional pollen because of interaction between cytoplasm and nuclear genes. Since CMS does not affect 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. The application of CMS/Rf system has proved to be an effective means to produce commercial  $F_1$  hybrid seed for many crops [1]. 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* L.) as a male parent to a wild species *G. harknessii* [2]. This system is involving cytoplasmic male sterility (A), maintainer (B) and restorer (R) line, has been widely used to develop hybrid rice varieties [3, 4]. 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 [5,6]. The use of this CGMS source is limited for want of potential restorers. In case of cytoplasmic male sterility system the abnormality of disintegration before pollen mother cell undergo meiosis has been reported by Khadi *et al.* [7]. Whereas in case of genetic male sterility system the abnormality is post meiotic. Therefore, the scope of CGMS system will be greater if more number of divergent restorer lines are identified. It was thought worthwhile to evaluate these CGMS lines for their potentiality as parents for hybrids and also to identify suitable restorers from the *G. hirsutum* accessions available in the gene bank.

*Gossypium harknessii* Brandagee (D2-2) which is a diploid ( $2n = 26$ ) was used as female by Meyer [8] to transfer *G. hirsutum* genome in the cytoplasm of *G. harknessii*. The resultant triploid was made hexaploid

( $2n = 78$ ) using colchicine. Male sterile tetraploid plants were recovered from cross between hexaploid and tetraploid. Seven cytoplasmic genetic male sterile (CGMS) lines, HS 6 CMS, K 34007 CMS, H 777 CMS, LH 1134 CMS, Bikaneri Narma CMS, SH 2379CMS and CSH 25 MCMS were developed at Central Institute for Cotton Research, Regional Station, Sirsa using IH 76 carrying *G. harknessii* cytoplasm by back cross breeding. To identify the restorer lines, the CMS lines were crossed to nine *G. hirsutum* male parents viz., CIR 15, CIR 23, CIR 39, CIR 47, CIR 51, CIR 54, CIR 56 CIR 58 and CIR 70 selected from the germplasm collections during 2001. Sixty-three crosses along with conventional check hybrid 'LHH 144' were grown in replicated trial of two row plot of 5.4m length having  $100 \times 45$  cm spacing between row to row and plant to plant during the *Kharif* 2002. On the basis of pollen dehiscence plants were classified as male fertile or male sterile. It was also confirmed in the laboratory with 1% acetocarmine test.

Out of 63 CMS hybrids, 25 (39.7 %) were fertile as CIR 23 and CIR 70 could restorer fertility in the 14  $F_1$ s and CIR 15 could restore the fertility for 6 CMS and CIR 47 for 5 CMS lines. (Table 1). The

**Table 1.** Fertile/sterile behavior of the *Gossypium* germplasm lines on seven different CMS lines

Female parent	Male parent								
	CIR 15	CIR 23	CIR 39	CIR 47	CIR 51	CIR 54	CIR 56	CIR 58	CIR 70
HS 6 CMS	F	F	S	F	S	S	S	S	F
K 34007 CMS	F	F	S	F	S	S	S	S	F
H 777 CMS	F	F	S	F	S	S	S	S	F
LH 1134 CMS	F	F	S	S	S	S	S	S	F
Bikaneri Narma CMS	F	F	S	F	S	S	S	S	F
SH 2379 CMS	F	F	S	F	S	S	S	S	F
CSH-25 M CMS	S	F	S	S	S	S	S	S	F

F = Fertile, S = Sterile

results revealed that 2 pure lines CIR 23 and CIR 70 acted as fertility restorer for all the 7 CMS lines. The *G. harknessii* cytoplasm of CMS lines HS 6CMS, K 34007 CMS, H 777 CMS, Bikaneri Narma CMS, SH 2379 CMS CSH 25 MCMS, LH 1134 CMS and CSH 25 M CMS may be same but there were differences in fertility restoration. Weaver and Weaver [9] also observed that a single gene, probably expressing partial dominance, controls fertility restoration in cytoplasmic male sterile cotton.  $F_1$  hybrid population showed a wide range of male fertility expression. Pollen production in the  $F_1$  (heterozygous for the restorer gene) was found to be much more variable and influenced by environmental conditions than in the homozygous parent. Different flowers on the same plant varied in fertility from practically sterile to highly fertile. Similar results were also obtained by Serieys [10]. He obtained partial restoration fertility and full fertility restoration by wild male parents on two different CMS lines of Sunflower having the same cytoplasmic background. Tuteja et al. [11] also reported that a maintainer of one CMS sometimes could not maintain the sterility of other CMS line, having the same cytoplasmic background. Seasonal influence on pollen production was also reported by Marshall et al. [12].

Two deleterious characters 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 [9]. Dutt et al. [13] also reported that sterile cytoplasm has detrimental effect on yield traits in hybrid cotton but this detrimental effect could be overcome by using better CMS lines and restorers with high restorability. 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, 14]. However no such deleterious effects were observed in the present study. The restorer lines CIR 23 and CIR 70 identified from the present study should permit the commercial production of completely fertile  $F_1$  cotton hybrids.

## References

1. Williams M.E.L.C.S. III. 1992. Molecular biology of Cytoplasmic Male Sterility, pp. 23-51 in Plant Breeding Reviews, edited by J. Janick. John Wiley and Sons, Inc., New York.
2. Meyer V. G. 1973. Fertility restorer genes for cytoplasmic male-sterility from *Gossypium harknessii*. Beltwide Cotton Prod. Res. Conf. Proc., p 65.
3. 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. Japan J. Genetics., **44**: 149-156.
4. 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.
5. Meyer V. G. 1965. Cytoplasm effects on anther numbers in interspecific hybrids. J. Hered., **56**: 292-294.
6. Meyer V. G. 1975. Male sterility from *Gossypium harknessii*. J. Hered., **66**: 23-27.
7. Khadi B. M., Katageri I. S. and Nalini A. S. 1994. Histological basis of CMS in Reba 50A cotton genotype. J. Indian Soc. Cotton Improv., **19**: 25-28.
8. Meyer V. G. 1971. Cytoplasmic effect on anther number in interspecific hybrids of cotton. J. Hered., **62**: 77-78.
9. 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.
10. Serieys H. 1999. Identification, study and utilization in breeding programme of new cms crosses. FAO Progress Report (1996-1999). Heli, **22** (Special issue): 103-107.
11. Tuteja O. P., Kumar Sunil and Singh Mahendar. 2004. Identification of new fertility restorers based on cytoplasmic male sterility system in cotton (*G. hirsutum*). International Symposium on Strategies for Sustainable Cotton Production — A Global Vision 1. Crop Improvement, 23-25 November 2004, University of Agricultural Sciences, Dharwad, Karnataka, India, pp 107-111.
12. Marshall D. R., Thomas N. J., Nicholls G. H. and Patrick C. M. 1974. Effect of temperature and day length on cytoplasmic male sterility in cotton (*Gossypium*) Aust. J. Agric. Res., **25**: 443-446.
13. Dutt Yagya, Wang Xue De and Nirania K. S. 2004. Effect of sterile cytoplasm on lint yield and contributing traits of  $F_1$  hybrids in cotton (*Gossypium hirsutum* L.). International Symposium on Strategies for Sustainable Cotton Production- A Global Vision 1. Crop Improvement, 23-25 November 2004, University of Agricultural Sciences, Dharwad, Karnataka, India, pp. 216-219.
14. Sheetz R. H. and Weaver J. B. Jr. 1980. Inheritance of a fertility enhancer factor from Pima cotton when transferred into upland cotton with *Gossypium harknessii* Brandegee cytoplasm. Crop Sci., **20**: 272-275.