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INDUCTION OF MALE STERILITY IN SESAME (SESAMUM INDICUM L.)

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ABSTRACT

Hybridization and mutagenesis have been applied in India since the 1970's in order to improve several seed crops as wheat, soybean, maize, groundnut, sunflower, rice, sesame, cowpea, moth bean, etc. Many mutant varieties were approved as national varieties and some promising regional lines through mutagenesis. Though exhaustive work has not been done on the male sterility systems and identification of restorers in *Sesamum*, there are few studies on genetic male sterility (GMS) and cytoplasmic male sterility (CMS) and also reports of male sterility during the study of cytogenetics or crossability using wild species in sesame. The possible applications of genetic male sterility (GMS) in plant breeding are reviewed and discussed. The basic contribution of GMS is that it provides a means of genetic emasculation which can be applied for the massive production of hybrids. There are two main fields of application, the production of genetic variation into crop varieties. Induction of genetic male sterility system coupled with natural honey bee activity can provide an effective tool for hybrid seed production in sesame. An attempt was made to induce genic male sterility system through a chemical mutagenesis. Two male sterile plants have been developed and maintained through sib mating. The male sterility system was found to be unstable in sesame. Stable male sterile lines are to be selected through repeated selection.

Keywords: sesame, mutagens, male sterility.

INTRODUCTION

Male sterility may have multiple causes. It can result from adverse growth conditions, from diseases, or from mutations. Naturally occurring genetically male sterile plants in hermaphrodite species generally maintain fully normal female functions. The phenotypic manifestations of male sterility are very diverse from the complete absence of male organs, the failure to develop normal sporogenous tissues (no meiosis), and the abortion of pollen at any step of its development, the absence of stamens dehiscence or the inability of mature pollen to germinate on compatible stigma. Use of artificial mutagenesis in breeding programs leads to increased variation in several economically important traits in crop plants. In sesame, improvement in different vegetative (Sengupta and Animesh, 2004; Hoballah, 1996) as well as biochemical (Savant and Kothekar, 2011; Savant et al., 2009; Cigdem et

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al., 2007) traits has been reported following artificial mutagenesis. In these studies substantial variation was reported for yield related traits and other agronomic and genetic traits (Sengupta and Animesh,2004; Hoballah, 1996). Hybrid breeding is one of the best methods to increase productivity in sesame. Though heterosis was reported as early as 1945 by Pal, as of today, the commercial exploitation of heterosis is not feasible due to the lack of economic means of hybrid seed production.

The male sterility system becomes an effective tool for hybrid seed production. The existing genetic male sterile (GMS) materials are not good. Their male sterility rates are low (less than 50%) and their agronomic characters are not ideal. Induction of mutation is an efficient method for creating new breeding materials, so research was carried out to create new male sterile breeding material that would be ideal and be used for heterosis breeding directly. Earlier male-sterile (MS) mutants (Kaul, 1998; Nilton *et al.*, 2005) and their cytological studies (Nilton *et al.*, 2005; Bione*et al.*, 2002) have been reported in many species of higher plants as the result of both spontaneous and induced mutations. Different chemicals affect male sterility in sesame by using different chemicals.

MATERIALS AND METHODS

In present investigation healthy seeds of two varieties of sesame namely JLT-7 and Western-11were treated with different concentrations of EMS (ethyl methanesulphonate) and SA (sodium azide) for two hours and sown in a randomized block design with three replications at the Plant Breeding Farm Dr. Babasaheb Ambedkar Marathawada University, Aurangabad during 2010. All M1 plants were selfed and harvested individually. In M2, the seeds from the single plants were grown in progeny rows and the mixed seeds of each dose treatment were grown in a bulk. The male-sterile plants could not be distinguished from fertile ones until capsule formation, when a low number of capsules with reduced size or their total absence under isolation were observed on green plants. So for early detection of sterile plants, at flowering stage, anthers of each M2 plant were observed carefully and their pollen studies were made with the 1% acetocarmine method.

All the M2 plants were tested at flowering stage for pollen sterility by the 1% acetocarmine method and classified as male fertile and male sterile based on the pollen morphology and stainability. Fully developed and red stained pollen grains were treated as fertile while shriveled and unstained pollen grains were grouped as sterile. The process of microsporogenesis is studied by taking the paraffin sections of sterile and fertile anthers. The procedure outlined by Johansen (1940) is used to study microsporogenesis. Photomicrographs were taken from the permanent slides. Other vegetative characters of sterile plants were studied and compared with normal plants.

RESULT AND DISCUSSION

Among the two mutagens used, mutagen SA succeeded in inducing male sterility in two plants out of 12462 plants tested in M2 generation. In M2 generation only two plants of variety W-11 named as wms-1 and wms-2 were found to be completely male sterile. We examined microsporogenesis and the pollen grains of male sterile plants. In fertile anthers, early anther development showed dense cytoplasm whereas in steriles, they were vacuolated. In the late microsporogenesis, locules of fertile were filled with normal pollen grains (Plate -2) and few sterile pollen grains appeared in sterile anther sections (Plate -3). We examined that the normal type pollen grains were spherical, had exine walls with visible apertures through which the future pollen tubes could emerge. By contrast, pollen grains of male sterile plant exhibited shriveled and collapsed morphology. There was no significant difference in days to flowering and days to maturity in *male sterile plant*. The results showed that there was a general reduction in the values of all the biometrical characters except number of branches per plant as compared to control (Table 1). Reduction in the values of all the biometrical characters was also recorded by Koh and Heu, (1995) in rice. Moreover wms-2 male sterile mutant produced with increased no of small anthers. In wms-2 six anthers were found as compared four anthers in control of W-11 (Plate 1). The shape and size of the pollen grains was found to be reduced than control. The anther color of the male sterile plants from wms-1 and wms-2 was brown. The anther shape of all male sterile plants was flat and a little shorter than the normal anthers. Moreover, male sterile plants produced small flowers, small brown empty anthers containing sterile/no pollen. Meiosis was aborted in MS plants generating abnormal microspores. This implies that the MS gene affects whole stages of growth and development. MS phenotype was stable regardless of air temperature and day length.

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Agronomic character	Control	<i>wms</i> – 1	<i>wms</i> – 2
Days to flowering	39.16 ± 0.18	37.00	38.00
Days to maturity	79.16 ± 0.329	78.00	77.00
Plant height	95.33 ± 0.69	91.00	
Number of capsules per plant	66 ± 1.7	24	19
Length of capsule	2.18 ± 0.069	1.83	1.78
Number of Seeds per Capsule	65.66 ± 0.268	34.43	33.87
1000 Seed Weight (gms)	3.25 ± 0.06	1.34	1.54

Table 1. Agronomic character of male sterile plants.





Plate - II Microsporogenesis in male fertile anther

In wms-1 the size of some of the capsules reduced very much and their length measured up to 0.5 cm with increasing number of locules. Surprisingly ten locules are recorded in some of the capsules of wms-1 (Plate 1). It was interesting to note that on sib pollination, male sterile plants produce fewer capsules in comparison to open pollination. Further, sib mated progenies failed to produce equal numbers of male sterile and male fertile plants. The frequency of male sterile plants was always lower than the expected level¹⁵ indicating that induced ms genes for genetic male sterility system have not attained stability in their expression. Reviewing genetic male sterility, Gottschalk (1976) was of the opinion that male sterility could be caused by genes due to partial or complete transformation of stamens into carpels in many species. The gene action would lead to transformation of bisexual flowers of a species into unisexual ones. In strictly self-pollinated species these mutants were usually seed sterile with the exception of Triticum mutant. However, earlier Osmanet al. (1982) reported that the male sterility system was stable in all environments. Gamma ray induced male sterility has been reported in pepper by Daskalov (2001), in lentil by Srivastav and Yadav (2001) and in niger by Sujatha (2001). Prakashet al. (2001) reported that different chemicals affect male sterility system in sesame. In



Male sterile flower



Plate - IIIMicrosporogenesis in male sterile anther

sesame, induced male sterility was reported by several workers (Osman *et al.*, 1998; Yingde *et al.*, 1998; Ganesan, 1998) which had proved useful for the hybrid seed production (Parry *et al.*, 2001). Anitha and Ganesan (2003) succeeded in inducing male sterility with reduced flower size and small brown anthers in sesame whereas the anther developmental defects in male sterile mutant of *Arabidopsis thaliana* were studied by Paul *et al.* (1999).

The present results indicate that mutation breeding continues to be a powerful tool to develop heterotic hybrids using induced mutants and also to induce male sterile mutations.

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