## MALE STERILITY IN INDIAN MUSTARD (BRASSICA JUNCEA (L) COSS) II. Genetics and Cytology of MS-1

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Traditionally pure line cultivars have been bred in Indian mustard. The development of  $F_1$  hybrids and population improvement is imperative to sustain the generation of new high-yielding varieties. Large scale manual emasculation and crossing of species with perfect flowers is tough thus genetic sterilization is of special interest. So the efforts were made to identify some male sterilizing genes/cytoplasmic sources for possible use in commercial hybrid seed production.

## Material and Methods:

Few male sterile plants were found in the open pollinated population of a German introduction EJ-32, during winter, 1979. Each male sterile plant was sib pollinated with fertile plants of EJ-32, and also crossed with two fertile accessions, RLM 198 and BS 119. Open pollinated seeds were collected from individual sterile plants separately. During off season 1980, all the  $F_1$ 's and sibs were grown and selfed to produce F2 generation. In winter 1980, all the  $\bar{F}_1$ 's,  $\bar{F}_2$ 's and sibbed progenies were grown and plants classified for male sterility. Fertile segregants in F2 generation were selfed to produce F3 family seed. Plant to progeny rows were sown during winter 1981 and scored for segregating and nonsegregating families. For cytological studies, buds from sterile and fertile plants were collected, fixed, dehydrated and finally embedded in paraffin wax. Transverse sections, 10 micron thick were dewaxed hydrated and stained in bromophenol blue (Chapman, 1975) and studied under light microscope.

## Results and Discussion :

Compared to fertile sibs, the male sterile plants were shorter, had more secondary branches, poor pod set, smaller pods with fewer seeds and lower seed fertility. The flowers of fully male sterile plants, in general had smaller bud, narrow petals,

abnormal stamens with short filaments and poorly developed anthers (Fig. 1a). Anthers had little or no fertile pollen. Female fertility (calculated as seed set on artificial sibbing expressed as the percent of total number of ovules per flower) was normal.

Genetic studies: The complete fertility in  $F_1$  suggested the dominance of fertility over male sterility. The overall segregation from  $F_2$  and  $F_3$  was compatible (P = 0.02-0.03) with the null hypothesis of monogenic recessive inheritance (Table 1), except in one  $F_2$  population from open pollinated seed at 1 per cent level of significance. This can be attributed to misclassification errors as the possibility of digenic inheritance was excluded by the progeny analysis of fertile  $F_2$  plants. Heterogeneity among  $F_2$  families was non-significant.  $F_3$  family analysis (Table 2) supported the hypothesis of recessive inheritance. Results suggest that male fertile individuals were 'MSMS' or 'MSms'while'ms ms' represented the sterile genotype.

Cytological studies: Deviations in the microsporogenetic developmental sequence started only after the release of quartets. The first abnormality observed in MS-1 was the weakly stained nuclei of mononucleate microspores (Fig. 1d). Nuclear condensation was apparent during early microspore mitosis. Chromatin condensation resulted in darkly stained mass that usually degenerated (Fig. 1e, s). Some variously sized dark masses were also observed in few microspores which was followed by vacuolation. These vacuoles increased in size thereby crushing the chromatin material (Fig. 1f). Empty microspores with normally developed walls were present in some mature anthers (Fig. 1g).

Post meiotic degeneration of tapetum was identical to fertile anthers till late mononucleate microspore stage. Subsequently there was rapid depletion of cytoplasmic contents, and cells became vacuolated with weakly stained nuclei (Fig. 1g). Ultimately a mass of hollow, flattened, unstained and twisted cell capsules was left. Degeneration of male gametophyte and tapetum due to vacuolation has also been reported in wheat (Simonenko, 1973) and cotton (Murthi and Weaver, 1974). In sweat pea, Childers (1952) ascribed such vacuolation to nutritional deficiency. A disruption in the biochemical link between tapetumthe nurse tissue and the developing microspores cannot be ignored. Evidences are available to show the transfer of macromolecules from tapetum to the sporogenous tissue (Heslop-Harrison, 1972). Such a link is important for suppression of tapetal DNA synthesis during meiosis, and translocation of DNA break down products and pollen wall precursors from tapetum to young microspores (Mian et al. 1974). Scanning electron microscope analysis of pollen grains of sterile pollen grains revealed a manifestation of retarded growth. Basic exine pattern, however, remained the same (Fig. 1h-i).

Cytological studies have shown consistent and complete male sterility of MS-1. The visible markers, narrow petals and small transluscent anthers provide an efficient way of classifying these plants under field conditions. Female fertility seems to be normal. Thus, this system of male sterility has a potential for use in hybrid seed production in general, and for population improvement programmes in particular.

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