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**Kugashiya KG**
Assistant Research Scientist, Department of Seed Technology, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Banaskantha, Gujarat, India

**HS Patel**
Ph.D. Student, Department of Genetics and Plant Breeding, C. P. College of Agriculture, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Banaskantha, Gujarat, India

**Chaudhari PR**
Assistant Research Scientist, Department of Seed Technology, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Banaskantha, Gujarat, India

**Desai TA**
Agriculture officer, Department of Seed Technology, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Banaskantha, Gujarat, India

**Vaghela PO**
Agriculture officer, Department of Seed Technology, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Banaskantha, Gujarat, India

**Corresponding Author:**
**Kugashiya KG**
Assistant Research Scientist, Department of Seed Technology, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Banaskantha, Gujarat, India

# Breeding for CGMS system in mustard (*Brassica juncea* L. Czern & Coss): A review

**Kugashiya KG, HS Patel, Chaudhari PR, Desai TA and Vaghela PO**

**Abstract**
The production of rapeseed mustard has undergone extraordinary change since last two decades. The requirement for oils and fats will be higher in view of ever increasing population in future. The practical option only left is the vertical increase in yields. Hybrid technology has shown to be a promising in this regard. A number of diverse CMS systems were developed. Evaluation of available CMS sources for flowering and seed yield traits for development of seed production technology of the hybrids and for the researchers to exploit the heterosis would be essential**.** In this study, development of different kinds of male sterility in different *Brassica* species will be reviewed.

**Keywords:** *Brassica juncea,* CMS lines, flowering, seed yield

## Introduction
The Brassica genus contains more than 3000 species of which forty are of economic importance. Most of the species are used as oilseed crop, vegetable crop and some as forage crops. Cultivated Brassica can be broadly divided into two distinct groups *viz*. 1) Oil seed type: Rape seed and Mustard 2) Vegetable type: Cabbage, Cauliflower and Turnip. Among the all Brassica species, major area is under *Brassica juncea* (2n=36) which contributes about eighty per cent of the total production in the country. Mustard seeds contain 40 to 45 per cent oil and 38to 41% protein. The development of high-yielding hybrid varieties has an important role to fill the gap between food production and human population. Heterosis and combining ability studies in *Brassica* species lead to the development of the hybrid varieties that will produce better seed yield, oil content, protein content and canola type traits for good human health. Cytoplasmic male sterility (CMS) is one of the most important approaches to utilize the heterosis in *Brassica* species. The characterization and discovery of novel male sterile cytoplasm is always the base for heterosis breeding of rapeseed.

## Floral biology
- The inflorescence is a corymb raceme.
- Petals are narrow and pale yellow to cream in colour.
- Sepals are united and green which turn yellow before falling.

## Androecium
- The stamens are six and tetradynamous. (tetra - four, dynamis – strength).
- Out of six stamens, four inner stamens are with long and two outer stamens are with short filaments.
- In the bud stage the immature stamens are always below the stigma.
- Just before the opening of the flower, the four stamens which have long filaments carry the anthers up to the stigma and the anthers are introse. (Anthers which have their face towards the periphery of the flower is called extrose anthers. When the face of anther lies towards centre of the flower, it is known as introse anther).

## Gynoecium
- The flower bears a hypogynous, syncarpous ovary (Syn – together or united, Karpous – fruit or ovary), which is bicarpellary with a very large number of ovules and parietal placentation. Before the flower opens, the style often increases in length.

**Male Sterility**

- Male sterility refers to a condition in which non functional pollen grains are produced in flowering plants.

**Male sterility types**

1. Genetic male sterility: A) PGMS B) TGMS C) TrGMS
2. Cytoplasmic male sterility 3) Cytoplasmic Genetic male sterility 4) Chemical induced male sterility.

**Source/origin of different CMS System**

1. **Autoplasmic CMS:** Lines are the result of mutations (Spontaneous or induced) in the native mitochondrial genome of a cultivated species.
2. **Alloplasmic CMS:** Lines are produced by transferring the cytoplasm from an alien/allied species in the background of a cultivated species.

**Development of *Moricandia arvensis* – based CMS and fertility restoration system in mustard *Brassica juncea* [Prakash *et al.*, (1998)] [7].**

Synthesize a steady CMS system *in B. juncea* by introduction of its nucleus in the cytoplasm of a wild species *Moricandia arvensis*, followed by repeated backcrossings of the resultant somatic hybrid *Moricandia arvensis* + *B. juncea*. *M. arvensis* is a wild plant in the Mediterranean area. The introgressing of genetic information for fertility restoration *from M. arvensis to B. juncea* through homoeologous recombination, and have thus perfect a cytoplasmic male sterility-fertility restorer system created. The resultant somatic hybrid *M. arvensis* (2n=28 MM) + *B. juncea* (2n=36, AABB) carrying the mitochondrial (*mt*) and chloroplast (*cp*) genomes of *M. arvensis*, was repeatedly crossed to cultivated variety of *B. juncea* cv Pusa Bold.

**Overcomes of leaf Chlorosis by chloroplast substitution in a *Moricandia arvensis* based cytoplasmic male sterile *Brassica juncea* [Kirti *et al.*, (1998)] [9].**

The development *B. Juncea* male sterile line from *Moricandia arvensis* cytoplasm showed developmental / floral abnormalities include leaf chlorosis of various degrees. To achieve chlorosis rectification it has resorted to an additional cycle of protoplast fusion, involving the chlorotic male-sterile (*Moricandia*) *B. juncea* and green fertile *B. juncea*. A resultant green male-sterile intergeneric cybrids carrying mitochondria of *M. arvensis* and chloroplasts of *B. juncea.*

**Stigma receptivity evaluation for CMS lines of *Brassica juncea.* (Chakarbarty *et al.* (2003) [5].**

For checking of stigma receptivity of seven CMS lines of *B. juncea* in Pusa Bold (PB) background was sownn in winter the season of 2002-03 in the field of Indian Agricultural Research Institute, New Delhi. The Pusa Bold plants were sown along with the CMS lines as pollen parent. Male sterile plants of CMS lines were identified and tagged. Unopened flower bud likely to be open in the inflorescence was covered with butter paper bag. Immature buds and open flower were removed. Flower was pollinated manually from the day it opened. Pollination were done up to 10 days in 2002-03 on different sets of flowers already covered before opening with fresh pollen of Pusa Bold. After pollination the flowers were covered. The experiment was conducted on 5-10 inflorescences with 5-15 developed buds in each inflorescence of the CMS lines in peak flowering period. Data were recorded on number of pods set with seeds per siliquae on the pollinated flowers of each sterile line. Oxyrrhina CMS lines have higher stigma receptivity up to ten days among the all the CMS Line.

**An assay of CMS lines of Indian mustard [*Brassica juncea* (L.) Czern. & Coss.] For flowering and seed yield traits [Shiv and Chakrabarty (2002)] [10].**

An experiment was conducted to estimate the seven cytoplasmic male sterile (CMS) lines of *Brassica juncea* in Pusa Bold background with the variety Pusa Bold for seed yield and flowering characters. The results revealed that Pusa Bold variety was the earliest in flowering. So, problem of non-synchrony in flowering of CMS lines and Pusa Bold was not observed. The *Erucoides* CMS lines was the earlier in bud initiation and first flowering while *Moricandia* (chlorotic) was the last among all the CMS line. This result might be due to the interaction differential of the nuclear cytoplasm of these two lines in general nuclear background of Pusa Bold. The Pusa Bold and CMS lines were at par in flowering duration except for *Moricandia* (chlorotic). No significant difference was observed in termination of flowering among all the CMS lines except *Moricandia* (chlorotic). The same was true for flowering duration.

**Detection of AFLP markers linked to the male fertility restorer gene of CMS (*Moricandia arvensis*) *Brassica juncea* [Ashutosh *et al.*, (2007)] [7].**

The Linkage analysis of Amplified Fragment Length Polymorphism and SCAR markers with the fertility restorer gene and with one another was determined by analyzing the co-segregation data. The used markers were found linked to the fertility restorer gene and mapped on one side of the *Rf* locus. The genetic distance between the marker and the *Rf* locus ranged from 0.6 to 2.9 cM. The marker SCAR3 was found tightly linked to the *Rf* locus with a map distance of 0.6 Centimorgan.

***Brassica juncea* carrying cytoplasm of *Diplotaxis catholica*: genetics of fertility restoration [Pathania *et al.*, (2003)] [8].**

A Cytoplasmic Male Sterile line of *B. juncea* was developed by placing its nucleus in the cytoplasm of an alien wild species, *D. catholica*, through sexual hybridization followed by repeatedly backcrossing. The restorer gene identified for this CMS in the progeny derived from the somatic hybrids of *D. catholica* + *B. juncea.* The number of locules in the silique was also governed by nuclear cytoplasmic interaction and is linked to CMS line. The *D. catholica* cytoplasm based CMS line has petaloid anther and shows low female fertility and gynoecium abnormalities.

***Diplotaxis erucoides*-induced cytoplasmic male sterility in *Brassica juncea* is rescued by the *Moricandia arvensis* restorer: genetic analysis. [Bhat *et al.*, (2006)] [3].**

An alloplasmic cytoplasmic male sterile (CMS) line of *Brassica juncea* was developed by repeated backcrossing with sexual hybrid *Diplotaxis erucoides* $_x$ *Brassica rapa* with *B. juncea* cv. Pusa Bold. The CMS line was comparable to the *B. juncea* nuclear donor line for all morphological traits apart from male fertility.

**Genetics of fertility restoration:** For identification of a fertility-restorer genotype for the new CMS line, restorer lines for alloplasmic CMS *B. juncea* with *E. canariense, T. ballii* or *M. arvensis* cytoplasm were tested. Among all only *M. arvensis* restorer line was found to be effective in conferring male fertility to the *erucoides* cytoplasm based CMS plants and the fertility restoration was gametophytic and monogenic.

**An improved cytoplasmic male sterile (*Diplotaxis berthautii*) *Brassica juncea* [Bhat *et al.*, (2008)] [4].**
Development of an improved male sterile line of *B. juncea* based on cytoplasm of *D. berthautii* which shows better floral features and high female fertility. CMS flowers have well developed nectaries but anthers are slender, sterile and fail to extend to the level of stigma. Male fertility of the CMS (*D. berthautii*) *B. Juncea* was restored by restorer lines of CMS (*M. arvensis*) *B. juncea* and CMS (*D. catholica*) *B. juncea.* However, restorers of CMS (*T. ballii*) *B. juncea* or (*E. canariense*) *B. juncea* do not restore fertility to *D. berthautii*-based CMS line. The male fertility restoration was governed by single gene acting at the gametophyte stage.

**A new cytoplasmic male sterility system for hybrid seed production [Sodhi *et al.*, (2006)] [11].**
The development of a novel CMS system in *B. napus* (named '126-1') and its transfer to *B. juncea* which could be used for hybrid seed production in this crop. CMS '126-1' was initially identified as a spontaneous CMS in a microspore derived doubled haploid population of *B. napus* and was then subsequently transferred to *B. juncea* lines by inter-specific crosses.

**Comparative Study of Five CMS System of Indian Mustard *(Brassica juncea*) with their maintainer line, Based on floral Biological traits (Gautam *et al.*, (2011)) [6].**
The study on floral biological traits of freshly opened flowers were randomly selected from the main shoot of each of the male sterile system and their maintainer at the time when entire line was in full blooming condition and observation of flowers of five male sterility systems was recorded and concluded that all the male sterile line have smaller size flowers in comparison to their maintainer lines. On the basis of comparison of flowers parts as petal, anthers, stamens and carpel length was found that the male sterile lines were smaller in size than their maintainer line.

**Exploitation of heterosis by using cytoplasmic male sterility system in Indian mustard [Brassica juncea (L.) Czern & Coss.] (Ashutosh *et al.*, (2014)**
For exploitation of heterosis, two CMS lines were crossed with thirty genotypes used as testers of Indian mustard in line x tester mating design, resulting in 60 hybrids. These thirty two parents (2 + 30) and 60 crosses were used to estimate heterosis over two standard check varieties *viz.,* Maya (Standard variety) and Kranti (Standard Variety). The highest economic heterosis for seed yield was recorded in CMSMori x NPJ-135 (13.17 %) over Kranti followed by CMSNDRE-4 x PRE 2007-06 was 12.83 % over Maya. These two hybrids were also showed high magnitude of heterosis may be exploited for commercial hybrid seed production by the use of advance male fertility restorer lines.

**Conclusion**

- Availability of alien CMS systems in Brassicaceae crops raises the question of adverse effects associated with alien cytoplasm, because use of CMS for commercial hybrid development would require absence of any such effects, which could be due to plastid or mitochondrial genomes.
- The negative effect of incompatible plastids causing leaf chlorosis in CMS *B. juncea* lines with Ogura, *B. oxyrrhina*, *M. arvensis* cytoplasm, and chloroplast substitution as an approach developed to overcome this problem.
- In *D. catholica*-based CMS, a restorer gene was only partially effective, petaloid anther, multilocular silique, and poor female fertility.
- Another effect that could come into play in alien CMS systems is the introgression of fertility restorer gene. Many CMS lines not used for hybrid development because of non availability of suitable male fertility restorer lines.

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