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Molecular Basis of Male Sterility in Crop Plants

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Introduction

One of the most prominent tools of plant breeding is the production of F1 hybrid plants. Resulting from differences in parental gametes, F1 plants show increased vigor and productivity based on hybrid vigor. Hybrid varieties are produced by controlled crosses between two distinct inbred lines, the optimal combination of which has been ascertained by extensive crosses. Since positive effects based on heterosis are only observed in the F1 generation, hybrid seeds always have to be produced by the breeder. Farmers demand a uniform genotype of the seeds; therefore, self-pollination of the seed producing plant has to be excluded. In order to prevent self-pollination in some plants like corn, tomato and cabbage, emasculation of the female line by hand is required, raising costs and labor expenses of seed production. Commercialization of any hybrid crop can only be achieved if reasonably priced technical solutions to hybrid seed production are available. Hence, with the use of genetic mechanisms of male sterility we can reduce the cost of hybrid seed production and ensure the availability of hybrid seed within the reach of poor farmers. Thus, the knowledge of molecular mechanism behind male sterility systems in crop plants is foremost things for a breeder to exploit the heterosis in a successful manner.

Male sterility

Male reproductive development in plants involves several major developmental stages in series and along several cell lineage pathways, which include specification of stamen primordia, production of sporogenous cells, development of tapetum and microspore mother cells (MMCs), meiosis, formation of free haploid microspores, degeneration of tapetum and release of mature pollen grains. Arrest of any of these steps can result in male sterility (MS), the failure to produce or release functional pollen grains. The phenotypic manifestations of MS may range from the complete absence of male organs, abnormal sporogenous tissues, to the in-ability of anther to dehisce or of pollen to germinate on compatible stigma. Male sterility is defined as the failure of plants to produce functional anthers, pollen, or male gametes.

Phenotypic expressions of male sterility include any of following signs,

- Absence or malformation of male organs (stamens) in bisexual plants or no male flowers in dioecious plants.
- Failure to develop normal microsporogenous tissue- anther.
- Abnormal microsporogenesis--deformed or inviable pollen.
- Abnormal pollen maturation; inability to germinate on compatible stigmat.
- Nondehiscent anthers but viable pollen, - sporophytic control.
- Barriers other than incompatibility preventing pollen from reaching ovule.

Classification of Male Sterility

a. Phenotypic

- ✓ Structural male sterility: anomalies in male sex organs (or missing all together)
- ✓ Sporogenous male sterility: stamens form, but pollen absent or rare due to microsporogenous cell abortion before, during, or after meiosis
- ✓ Functional male sterility: viable pollen form, but barrier prevents fertilization (anther indehiscence, no exine formation, inability of pollen to migrate to stigma or other factors that affect fertilization --e.g. extended style, pollen is glued together so can't be released-soybean, peas)

b. Genotypic

i. Genic male sterility: Mendelian inheritance due to nuclear not cytoplasmic genes

1) Arisen as spontaneous mutants in most cases: - high frequency of mutation.
2) Identified in 175 species.
3) Few reciprocal differences observed.
4) ms (recessive--most) or Ms (dominant--few)
5) Many nonallelic genes are known in some species (e.g. 60+ in maize, tomato 55, etc.)
6) Recessive: single genes ~70%; multigenes~15 (monocot)-23 (dicot) %: rest is dominant mutants- 7 (dicot) 15 (monocot)%.

ii. Cytoplasmic male sterility: Non-Mendelian inheritance - cytoplasmic

1) Reciprocal differences observed.
2) N (normal) and S (sterile) cytoplasms.
3) Male sterility inherited maternally.

4) Not very common.

iii. Gene-cytoplasmic male sterility: Both nuclear and cytoplasmic genes are involved

1) N and S cytoplasm.
2) Restorer of fertility (*Rf*) genes–distinct from geneic male strility genes.
3) *Rf* requires to restore fertility in S cytoplasm. N *RfRf* = fertiles; S*Rf*- = fertiles; S
4) *RfRf* = male sterile.

Several key genes and networks that determine male reproductive development, including the differentiation of sporophytic, specification of tapetum and microsporocyte cells, and biosynthesis and regulation of sporopollenin and pollen exine development. Mutations in such genes often result in MS in different forms.

MS systems used in hybrid rice production

Commercialization of any hybrid crop can only be achieved if reasonably priced technical solutions to hybrid seed production are available. In rice, hybrid seed production was first attempted using chemical hybridizing agent in the 1970s, but this approach was no longer used after MS systems became available. In order for an MS system to be workable for hybrid seed production, it must meet the following prerequisites: (1) complete and stable MS during hybrid seed production; (2) no substantial negative effect on MS and hybrid plants; (3) ability to multiply MS seeds through an intermediate genetic line (maintainer) or under particular environmental conditions; (4) ability to fully achieve fertility in hybrids. Therefore, although a number of MS systems have been generated during the past 40 years, only those that met these requirements were adopted in hybrid production. So far, two distinct systems have been utilized in hybrid rice production: cytoplasmic male sterility (CMS) and environment-conditioned genic male sterility (EGMS).

CMS systems

WA-CMS lines are the most widely deployed lines in hybrid rice production. Pollen abortion in WA-CMS occurs relatively early during microspore development, mainly at the uninucleate stage, resulting in amorphous aborted pollen grains. The pollen abortion is determined by the genotype of sporophytic tissues, not by the genotype of the pollen itself. That is, aborted pollens are only produced in plants with homozygous *Rf* (restorer of fertility) gene (s) and CMS factor(s), but not in plants that are heterozygous at the *Rf* locus (Figure 1, pollen fertility of F1 plants). All other CMS types of Indica rice, except for HL-CMS, are similar to WA-CMS and are classified as WA-CMS-like types.

EGMS systems

The other MS system that is widely used in hybrid rice breeding is the EGMS system, which includes the photoperiod-sensitive genic male sterility (PGMS) and temperature-sensitive genic male sterility (TGMS) lines. PGMS lines are male-sterile under natural long day conditions and male fertile under natural short-day conditions, whereas TGMS lines are sterile at high temperatures and fertile at lower temperatures. The majority (>95%) of the EGMS lines utilized in hybrid rice production in China were derived from three independent progenitor lines, i.e., PGMS line Nongken 58S (NK58S) and TGMS lines Annong S-1 and Zhu 1S. Some lines, such as Pei'ai 64S, are male sterile under both long day and high temperature conditions and are referred to as P/TGMS lines.

Approaches for development of male sterility

The specific mechanisms causing male sterility in plants vary from species to species and are subject to influence by environment, and nuclear and cytoplasmic genes. Male sterility may be permanent (heritable) or transient (CHAs). It can be induced by another culture somatic cell culture and somatic hybridization. Commercial exploitation of hybrid vigour in hermaphrodite crops is facilitated by the availability of CMS lines to affect pollination control. There are many ways to create Male sterility for instance: **A.** Cell cytotoxicity: *Barnase* – *Barstar* system, hormonal engineering, and patho genesis-related proteins. **B.** antisense RNA or RNAi construct. **C.** modification of biochemical pathways of flavonoids, jasmonic acid and carbohydrates. **D.** engineering mitochondrial and chloroplast genome

Conclusion

Genetically engineered male sterility provides tremendous opportunities to the breeders for enforcing pollination control in hybrid seed production systems. On the other hand, these systems have some disadvantages like availability of efficient gene construct, possible dispersion of transgene to other related species, availability of efficient transformation technique and very high initial investment. Apart from *barnase-barstar* system, no other system has reached the commercial stage. With the further researches on molecular biology of pollen development and improvement of biotechnology, the approaches creating male sterile lines using genetic engineering will become simpler, faster and more effective. They will be widely used to promote the heterosis utilization of vegetables and other crops and play more and more important roles in vegetable breeding.