Recent Developments in Studying Male Sterility and Heterosis Utilization of Oilseed Rape in Anhui China

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Anhui Province, located in the east of China (down reach region of Yangtze River) is a main agriculture area with important traditional oilseed rape producers. Annual sown acreage of oilseed rape in this province is one million ha, the largest in China. In order to match different cropping systems, *Brassica napus* and *Brassica campestris* are the main cultivated species of oilseed rape because Anhui Province is a transition area between southern and northern China both geographically and climatically.

We started quality breeding in the late 70's. The first low-erucic acid variety (B. napus) in China was registered in 1985 but it showed lower yield and susceptible to Sclerotinia sclerotiorum. It has been our top priorities that breeding for high yielding, double low (low erucic acid and low glucosinolates) and disease (Sclerotinia sclerotiorum) resistance (tolerance) variety since then.

1. CMS and Heterosis utilization in B. campestris

The male sterile plants of *B. campestris* were found in 1989 and sterile line was bred and studied. They belong to cytoplasmic male sterile according to the facts that there were maintaining and restoring genes among *B. campestris* varieties and there were no segregation in test cross and back cross generations. But male sterility was gradually turning to fertility after full flowering stage. Environmental effect on the variation of CMS line was conducted by treatments of artificial shading light, natural daylight, sown in fall and spring. Results suggested that there was no relation among the daylight lengths and the sterility variation, and there was also no significant effect of sowing dates on the variation, both the high temperature and accumulated temperature played an important role in the sterility variation

It was studied if it was possible to use the CMS line as two-line (male sterile line for FI hybrid seed production and inbreed line for male sterile line reproduction) by different sowing dates and picking flower stem. The results showed that the CMS line had longer duration of sterility during flowering period when sown later (in the beginning of November in Hefei) could be used as pollen control system to produce FI hybrid seed. The initial flowering stage of the CMS line could be postponed for more than 10 days to make the fertility duration of CMS line longer to reproduce male sterile line itself.

The ways of heterosis utilization in *B. campestris* were studies by using *B. campestris* inbreed lines which selfed continuously for 3 generations in 4 ways (a. bud pollination, b. sib-selfing, c. crossing between sister lines and d. spontaneous pollination). Crossses between the lines of every propagating way mentioned above were made to check F1 heterosis rate. Results indicated that the depression rates were different inbreeding way and the bud pollination was the lowest. The

higher depression rate parents had, the higher heterosis rate hybrids appeared. But heterosis also depends on general combining abilities of parents. The yield of the highest combination was 98.3% higher than their higher yield parent.

2. CMS and CMS three-way-cross hybrids in B. napus

Studies on stem rot (Sclerotinia sclerotiorum) resistance (disease avoidance) and tolerance in polima CMS lines of Brassica napus for 5 years. Results indicated that the average incidence and severity of the disease were 49.8% and 68.6% lower in sterile line 90A than in the maintainer 90B, respectively, and were 48.4% and 51.8% lower in sterile line 92A than in the 92B, respectively. The relative resistance of sterile lines had positive correlation with the smaller petal and lower rate of petal stuck on the leaf, and was, in fact, a sort of disease avoidance which could be inherited with the company of smaller petal controlled by sterile cytoplasm. Results from artificial inoculations indicated that disease tolerance between the sterile and maintainer was significantly different, the differences were caused by nitrogen metabolism and anthocyanidin content in the stem of plant. The higher content of 17 amino acids at green pod development stage and higher content of anthocyanidin at the end of flowering and green pod development stages in the sterile lines might play an important role in limiting expansion of oxalic acid toxin from the mycelium of Sclerotinia.

In order to overcome low hybrid seed production and trace pollen problems of sterile plants in polima CMS system, the trace pollen of sterile plants will also lead to selfing and reducing the seed purity when temperature fluctuates during hybrid seed production. Studies on heterosis comparison between CMS three-way-cross hybrids and single-way-cross hybrids, and probability of increasing yield of hybrid seed were conducted based on common parents in single-way and three-way crosses. The original male sterile lines crossed with new maintainers to create new single-way male sterile lines. The three-way-cross hybrids were produced through crossing the single-way male sterile lines with restorers. Results indicated that there were significant differences in individual plants yield among whole three-way-cross and single cross hybrids. The plant yield was higher in average three-way-cross hybrids than in average single cross. The yield Of 2 three-way-cross hybrids were even higher than all single-cross hybrids. The results also indicated that the agronomic characters of single cross male sterile line dominated that of original male sterile line with very significant level, the hybrid seed production had be increased by a big margin.

3. GMS and its use in hybrids breeding in Brassica napus

A series of genetic male sterile (GMS) lines (9012A, Y420A and 7024A) discovered in *B. napus* in the beginning of 1990's were used for the study. Results showed that 9012A, Y420A and 7024A were different from GMS line 6AB (with dominant gene epistiatic interaction, reported by Li S. et al in 1985 and 1988) in restorer-maintainer relationship, and all testers were restorers of the 3 GMS lines. Genetic studies revealed that their fertility was controlled by the interaction between recessive duplicate sterile genes (RDSG) and recessive epistatic inhibitor genes (REIG). Allelism testine indicated that 3 group's independent RDSG existed in *B. napus*. In addition, 2-3pairs of independent REIG are likely in *B. napus*. The interaction between REIG and RDSG was non-specificity, any 1 pair of REIG inhibited any 1 group RDSG, and 9012A involved 1 group RDSG and 1pair of REIG, Y420A

involved 1 group RDSG and 2 pair of REIG, 7024A involved 2 group's 1 pair of REIG. Based on this principle, temporary all maintainer (TAM) with 1 group homozygous RDSG and 1 pair of homozygous REIG have be bred, and a RGMS 3-line system (3LS) including homozygous double-purpose sterile line (DSL), TAM and restorer have been used for producing hybrid seed.

Several double-low (low-erucic acid and low-glucosinolates) hybrids have been bred through the GMS system. The 2 double-low hybrids C022 and 5C21 have passed the provincial and national official trials. The average yield of C022 was 7.2 % higher than CK Qingyou No.2 (high erucic acid and high glucosinolates hybrid) in 2 years adaptability tests, and 16.4% higher than CK in yield potential tests, and was 16.3 % higher than CK Z.Y.821 (high erucic acid and high glucosinolates normal variety) in national adaptability tests and was 18% higher than CK in yield potential tests. C022 has been registered and commercialized in China. Experiments results also indicated that the GMS system has some advantages over currently two sorts of GMS systems (one is controlled by two pair of dominant Ms genes and the other is controlled by two pair of recessive duplicated genes) It is convenient for this system to operate for hybrid seed production and easier to gain higher hybrid seed than any other GMS and CMS systems to do so for in China. It will be maximum for the system to improve F1 yield production of hybrids as restorer is easy to be found. Results also showed that the system is excellent to incorporate disease (Sclerotinia sclerotiorum) resistance (tolerance) into a variety.