

Breeding of the POL-CMS line BE23A in *Brassic napus*

CHEN Weijiang, LI Mei, FAN Lianyi

Hunan Crops Research Institute, Changsha, 410125, China Email: 93302@163.com

Abstract

Pol-CMS Line BE23A was bred by the Pol-cms 254A as sterile resource and the interspecific hybrid offspring (*B. napus* × *B. campestris*) as a new maintainer material through backcross and testcross for several generations. Some interspecific characters such as short plant, big petal and full nectary has been found which is useful for hybrid seed production. This bred line has stable male sterility with thin and long bud, shriveled anther and slow in reaction to low temperature. Over 95% purity has been gotten by this sterile line for commercial hybrid seed production in natural condition. Through the press selection, it is with double low quality and good economic characters. The combinations completed by BE23A were promoting yield 5.3%-19.5 than those completed by original sterile line 254A when the same restorer lines were used.

Key words: Pol-CMS, interspecific crossing, hybrid heterosis, *Brassica napus*

Introduction

Several of methods are used for hybrid heterosis in rapeseed but the cytoplasmic male sterility is the most important one for commercial hybrid seed production. In China the widely used CMS system is Pol-CMS. Use this system four varieties had been bred by our institute. Those varieties can promote the yield to 10%-20% generally compare the normal varieties and now the utilizing area is nearly a million hectare a year.

Plo-CMS system was built by Fu.T.D in 1990. Compare to the Ogura CMS, it is expressed sensitive to temperature in male sterility. But in practically using, the temperature sensitive problem can be relieved by change the sowing time or location, for example on south of China for propagation and north of China for hybrid seed production (Fang liangyi.2003). In this way it can adjust the blooming time for hybrid seed production under relatively high temperature and obvious enlarged scale of the Pol-CMS system utilization. But using this method can not be solved the phenomena of sensitive problem completely, so more stable Pol-CMS line is still needed. In this paper we used a interspecific hybrid material (*B.napus.L* × *B.Campestris*) as maintain resource and bred a new stable Pol-CMS line B23A. This new line has stable male sterility, double low quality, excellent yielding characters and strong resistance to diseases which is a very valuable material in hybrid heterosis utilization.

Breeding process of the Pol-CMS line B23A

A Pol- CMS line 254A was used as the female to be crossed with an interspecific intermediate material BE23 (*B. napus* × *B.campestris*) in 2000. And its offspring population was found the sterility segregation from semi-sterile to highly sterile degree. This phenomenon indicates that the material BE23 belonged to the maintaining type but affected by different minor gene which made the sterility separating in the offspring. This kind of material caused the selecting become possible.

From 2001 on, two methods were used to select the stable sterile line and its corresponding maintainer. One was initial screening by testcross in single plant pairs. That means that the same female plant was crossed with several different male parents. Completely sterile plants in offspring of segregation were selected to backcross respectively with different plants of BE23 and the desired male plants were selected by compare the offspring sterility for the next backcross generation. Another was two-way testcross in the late period of selection. According to branch number, each 2-3 desired MS plants selected from the initial screening were crossed respectively with each 2-3 corresponding maintain plant.

In the initial screening, sterile plants whose stamens in the location 1/4 lower than the pistils as highly sterile plants (HS) and those without any pollens as completely sterile plants (CS) were determined. Four combinations had HS plant over 50% and CS plants over 40% among the 11 backcross combinations in 2001. Five combinations had HS plants over 30% and CS plants over 60% among 12 backcross combinations in 2002. However with the method of the two-way testcross in the same year, 4 combinations reached over 90% in CS plant among 15 testcross combinations. Through further selection, the stable male sterile line BE23A and its maintaining line BE23 were successfully bred. It is indicated that it would be a useful method for initial screening at the early stage and two-way testcross at the late stage. Because both the desired sterile and maintain plant may be selected at the same time in the two-way testcross which will be accelerated the breeding program.

Male sterility performance in Pol-CMS line BE23A

Resourced from Pol CMS system, BE23A has the same genetic properties as others from the system in their relationship between fertility restoration and sterility maintenance. Its sterility is controlled by the Pol cytoplasm and a pair of major gene with multiple pair of minor gene sensitive to the low temperature. When it crossed with restoring line, the F₁ will restored the fertility, which is expressed by their completely fertile population. But when it crossed with ordinary varieties the F₁ will be expressed semi sterility because the latter carry no restoring gene and still influenced by minor gene. As for the maintaining

line BE23, it was origin from the interspecific cross and was bred by directive selection which decreased the effect of the minor genes to a very low degree, thus maintaining the sterility effectively. This function of maintain sterility is from *Campestris* gene or from the interspecific interaction still need to identify.

BE23A floral organ structure is a little different from that of its initial sterile resource 254A, the flower bud turn to slender from triangle in 254A shape, petals to ellipsoid and semi-slit from narrow long and all slit, and a little wider than the 254A's. BE23A has white, slender and empty anthers, comparatively big pistils and stigmas, and flat petals in dark-yellow color. Generally in pol CMS system, those CMS line with smaller petals and pistils usually have complete sterility but become bad in their economic characters. However BE23A has gone beyond this limit. Compared to the 254A its economical characters have been greatly improved. The floral structures of BE23A can be seen in Table 1.

Pol-CMS BE23A falls in type of another abortion. Under normal temperature at the flowering stage, it will reach high sterility degree, with its stamens abortive and wrinkled to white and without any pollen in it. But when it meeting the continuous low temperature below 12C for over a week, a few plants will turn to triangular shape in their anthers and produce a few pollens. After that it will turn to completely sterility in the late flowing stage. In hybrid seed production with the BE23A as the female, F₁ will not be greatly influenced in their fertility restoration because of the foreign pollen's competitive vigor, it much more than the BE23A's pollen. We used BE23A for hybrid seed production in the natural condition in recent years the rate of the restorations can be over 95%. it is much better than other Pol CMS line in our Institute and can promote 5-10% in fertile purity (table 2).

Biological properties of BE23A

Some interspecific characters have been found in BE23A such as deep green color and round shape in leaf, short plant in height, big petal and full nectary in floral organ. Also BE23A is an early-maturing CMS line, with initial flowering at early stage of March and finally flowering at early stage of April in Changsha area. And the F₁ competed with this sterile line will usually ripen 3-5 days earlier than the male parent even if it crossed with a late maturing restorer. In the field production, a few BE23A plant's initial flower buds will become yellow when meeting very low temperature. However, once it flowering, its flowers will turn normal without any yellowing and dying buds again. The BE23A has a strong vegetative growth, with vigorous seedling stage, erect leaves in dark-green color. Under average fertilizer application, it will have 10-12 green leaves before the winter and considerable resistance to cold during the wintering stage. From the next spring, it will grow stably and uniform which with medium plant height, developed first branch, compact pod grains and heavily 1000-grain weight (Table 3). At the same time its stem is sturdy that has good resistance to diseases and lodging. For practical application, the quality of BE23A also was gotten the desirable require by press selecting. The content of erucic arrived 1% in two generations and the glucosinolate arrived 35 μ mol/g in four generations.

As the BE23A has good economical character, the combination yield completed with it obvious increased. From 2004 on, 24 combinations were completed by BE23A, the F₁ increase yield 5.3%-19.5% over the combinations by 254A with the same restore parent. So BE23A not only improved the male sterile performance but also the yield character by interspecific cross. It has the fine prospect for commercial hybrid breeding.

Table 1. Floral structures of sterile line BE23A, maintain line BE23 and original sterile line 254A

Materials	Calyxes		Petals		Anthers		Pistils and Stamens	
	length	width	length	width	length	width	length	width
BE23	0.95	0.35	1.63	0.98	0.31	0.13	1.06	1.13
BE23A	0.90	0.25	0.27	0.77	0.15	0.06	0.95	0.36
254A	0.90	0.21	1.03	0.72	0.21	0.11	0.81	0.42

Note: Unite=cm

Table 2 Fertility restoration of the hybrid combinations in BE23A

Year	Plants tested	MS plants	F plants	Restoration rate %
2004	720	15	705	98
2005	1080	59	1021	95
2006	3600	113	3478	96

Table 3 Economical characters of sterile line BE23A

Year	Plant height (cm)	The first branches	Total pods	Pod's length	Grains	1000-grain weight (g)
2004	153.8	10.4	312.6	5.6	24.9	4.1
2005	157.0	10.7	349.4	5.4	23.8	4.2

Note: the date value is the average of 10 plants

References

- Fu, T.D., Yang, G. and Yang, X. (1990), Studies on "three line" polima cytoplasmic male sterility developed in *Brassica napus* L. *Plant Breed* 104: 115-120.
 Fan Lianyi, Chen Weijiang, Qu Liang. (2003), Seed production techniques of hybrid rape by sowing in spring in the North of China. *Hunan Agri Sci* 184: 12-15.