

# DEVELOPMENT OF COUNTERPARTS FERTILITY RESTORERS BASED ON STERILE CYTOPLASM UTILIZING DECAPITATION AND CHEMICAL CASTRATION OF SUNFLOWE PLANTS

L.K. VOSKOBOINIK; N.I. BOCHKAREV

The CMS source in sunflower was obtained by Leclerq in 1968 in an interspecific *Helianthus petiolaris* Nutt. x *H. annuus* Wensl. (1). This CMS source provided 100% sterility of plants, and difficulties arose in determining the sources of restorer genes. Leclerq was the first to find restorer genes in wild sunflower species but he failed to develop the initial breeding material possessing restorer genes.

M. Kinman (USA) was the first to find restorer genes in wild sunflower species but he failed to develop the initial breeding material possessing restorer genes.

M. Kinman (USA) was the first to find restorer genes within the population T 66006 which possessed hybrids with a wild annual sunflower in its pedigree (2). Later American scientists have found that wild annual sunflower species *H. annuus* and *H. petiolaris* possessed a high concentration of restorer genes (3).

Romanian breeders were the first to evaluate the world sunflower collection in the search of genes restoring fertility. Among the evaluated material restorer genes were found in the following varieties: Discovolante, Slovenska Siva, Mesohedesi, Blumington, Synthetic II (4). Bulgarian breeders have found restorer genes in the variety Pioneer of Siberia and in some local Bulgarian varieties-populations, Yugoslavian breeders have found restorer genes in the Soviet variety Mayak, in Argentine variety Klein and in some specimen of VIR collection: K-772, K-995, K-67, K-2138 (5). Fertility restoring genes were found in the variety Progress (developed at VNIIMK, Krasnodar) developed from the interspecific hybrid *H. tuberosus* x *H. annuus*.

The available data in literature show the low rate of restorer

genes in sunflower varieties-populations thus hampering the development of lines fertility restorers.

However, fertility restoring genes can be incorporated into the best male lines of hybrids, that is basing on the best lines the counterparts fertility restorers are developed.

There are several methods of developing such counterparts. The method of developing the counterparts on fertile cytoplasm is inconvenient as it requires a detection of restorer genes in each backcross generation.

Ekhart and Hadjinov (1954) proposed the method of developing fertility restorers based on sterility (6).

In this method of counterpart development the CMS detects the fertility restoring genes and the necessity in analyzing crosses is excluded. Maize plants have heterogamous flowers, i.e., crosses are performed without castration. To develop the counterparts restoring fertility in sunflower castration is necessary as the flowers are bisexual. However, this procedure is labour consuming one and has to be done during the limited period of time very early in the day from 4 to 6 a.m.

In order to facilitate and simplify the work of developing the counterparts fertility restorers a method was designed at VNIIMK enabling crosses with simultaneous development of a sterile counterpart and restoring counterpart (7).

According to this method the fertility restoring counterpart is developed basing on a simultaneously developed sterile counterpart.

The proposed method totally eliminates the necessity in manual castration though it is more time consuming (for 1-2 crosses) compared with that proposed by Ekhart and Hadjinov.

In order to shorten the period of time necessary to develop counterparts fertility restorers it is proposed to base on the method of crosses after Ekhart-Hadjinov.

$$\begin{aligned} & (Xc \times R) \\ & (Xc \times R) \times B \\ & / (Xc \times R) \times B / \times B \end{aligned}$$

According to the pattern of crosses the first cross is made between the CMS source (Xc) and the source of fertility restoring genes (R). We propose to start the crosses with source of fertility restoring genes possessing the sterile cytoplasm and the line B for which the counterpart fertility restorer is being developed.

(R x B)  
(R x B) x B  
((R x B) x B) x B

Thus, the period of developing the counterpart fertility restorer is shortened by one cross compared with the method by Ekhardt—Hadjinov and by two crosses compared with the method developed in VNIIMK. In cross I all female plants are fertile; in order to exclude manual castration of flowers during the bud setting stage the plants at this stage are treated with gibberellin, i.e. chemical castration of plants is made. Before flowering stage 6-7 plants are isolated in the source of restorer genes, and by the time of pollination the isolated plants are checked for sterility. The F<sub>1</sub> plants are heterozygous for fertility restorer gene. They are grown in one-row plots; the neighbouring line is the one for which the counterpart fertility restorer is being developed.

In the rosette stage all F<sub>1</sub> plants are treated with gibberellin, and before the opening of tubular flowers 6-7 plants are isolated. By the moment of pollination the plants are checked for sterility and thus 4-5 plants are left for crosses. In the first backcross progeny segregation for fertility is observed. The BC<sub>1</sub> seeds are planted in two-row plots neighbouring the line for which the counterpart is being developed. BC<sub>1</sub> plants in the seedling stage (the first pair of true leaves) are decapitated. Following the elimination of the growth point two shoots appear similar in development (Fig. 1). When the shoots reach the stage of rosette one of them is treated with a gibberellin solution (concentration: 0.0035%). As gibberellin somewhat increases the rate of development, for the gibberellin treatment one should choose the tiller which is behind the second one in development (Fig. 2). Before the opening of tubular flowers tillers treated with gibberellin are isolated. At least 15 plants are isolated for every crossing combination. Plants with restored fertility are chosen for backcrosses among gibberellin untreated control tillers. Plants having sterile flowers on the control, untreated tillers are screened off, i.e. bags are taken off the gibberellin treated tillers. For the needs of pollination plants are selected possessing sterile tillers following the chemical castration and control tillers with normal fertility. In spite of the fact that the head diameter of the developed tillers is almost two times less than in normal one-head plants, seed setting is rather high (134-729 seeds per plant) and sufficient for planting and growing the necessary number of plants for subsequent backcrosses. Thus, plant decapitation, chemical castration and crosses are made the following year.

Backcrosses should be effected until morphological identity of the fertile line and the counterpart restoring fertility is reached. As a rule, the initial line and its counterpart reach identity after 5-6 backcrosses. When phenotype identity of the line and its counterpart are reached the plants of the fertility restorer are selfed in order to shift restorer genes into homozygous state.

The proposed method of developing counterparts fertility restorers basing on sterile cytoplasm utilizing decapitation and chemical castration totally excludes the unproductive process of manual castration. Compared with the method of simultaneous development of sterile counterparts and fertility restoring counterparts this new method reduces the period of counterpart development by 1-2 crosses.

## REFERENCES

- (1) LECLERC, P., Cytoplasmic male sterility in sunflowers. Third International Sunflower Conf., US, 40. 1968.
- (2) KINMAN M., New development in the USA and state experiment station sunflower breeding programs. Fourth Int. Sunflower Conference, US, 1970.
- (3) FICK G., ZIMMER D. E., JIMÉNEZ G. D., BEDER D. A., Fertility restoration and variability for plant and seed characteristics in wild sunflower. Sixth Intern. Sunflower Conference, Romania, 1974.
- (4) VRANCEANU V., STOENESCU F. Surse de gene restouratoare a fertilitatii polenului la floarea-soarelui. Ann. Inst. Cerc. cereale Plante Tehn., Fundulea, Ser. C. 39, 1973.
- (5) SCORIC D., CUK L., MIHALJCEVIC M., MARICOVIC R., New sources of fertility restoration and dormy mildew resistance in sunflower. 8th Internat. Sunflower Conf. USA, 1973.
- (6) HADJINOV M., Line breeding and fertility restoration. Journ. Kukuruz, URSS, 1961, 1.
- (7) VOSCOBOINIK L. K., BOCHKAREV N. I., Development of counterparts fertility restorers in sunflower. Journ. Selekcija i Semenovodstvo, URSS, 1979, # 6.