Sergey V. Gontcharov*, Tatyana S. Korotkova, Natalya N. Goloschapova and Alexander P. Nesmyslenov **Shuttle breeding in sunflower lines development**

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Abstract: Shuttle breeding is a breeding system where generations undergo sequential evaluations at different locations, developed by N. Borlaug. Our study aims to demonstrate an opportunity of application shuttle breeding method in sunflower lines development and also to show its advantages and disadvantages. Experiments were mainly conducted at the Central Station (Krasnodar) of All-Russia Research Institute of Oil Crops (VNIIMK). Krasnodar region is situated in the Southern part of Russia near the Black Sea. Part of our work was made at Seed and Plant Improvement Institute (Karaj, Iran) and Experimental Station near Borozdjan (Busher Province of Iran). Released and experimental sunflower hybrids and lines of VNIIMK breeding were used as a material. The experimental design was randomized blocks with three replications. Each replication had four rows and two central rows were analyzed only to exclude the border effect. It is shown that the application of the shuttle breeding technique to sunflower was highly successful and results in elite line development.

Keywords: hybrid; line; pedigree; shuttle breeding; sunflower.

Introduction

Shuttle breeding – any breeding system where generations undergo sequential evaluations at different locations. The method was proposed by the prominent plant breeder N. Borlaug. He used it in wheat breading since 1945 (Borlaug 2007).

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The initial aim of the method was to speed up the breeding process by using extra generation per year. But results were much greater. The efficiency of selection significantly increased. Different biotypes look similar at one location differentiated at another one. Additionally, photo neutral wheat varieties were developed.

Shuttle breeding is widely used in wheat breading till now, for example at Omsk Agricultural University in Siberia, Russia as a part of the network of International Maize and Wheat Improvement Center (CIMMYT) (Shamanin 2009).

Despite the high effectiveness of the method, it is used rather rear for the other crop breeding. Probably the reason is evident organizing difficulties. The only organization, as far as I know, that put shuttle breeding at the core of the breeding program is the Eastern India Rainfed Lowland Shuttle Breeding Network (EIRLSBN). EIRLSBN developed 20 released rice cultivars and scores of elite lines for hybrid rice production in 20 years period (Mackill et al. 2013). This is a good example of international and interregional cooperation.

Our study aims to demonstrate an opportunity of application shuttle breeding method to sunflower lines development and also to show its advantages and disadvantages. Sunflower is one of the most important oil crops in the world and the main oil crop in Russia.

Materials and methods

Experiments were conducted at the Central Station (Krasnodar) of All-Russia Research Institute of Oil Crops (VNIIMK) from 1996 till 2019. Krasnodar region is situated in the Southern part of Russia near the Black Sea. Climatic conditions are very favorable here for sunflower production. Sunflower usually covers about 0.5 million ha in this region. Part of our work was made at Seed and Plant Improvement Institute (SPII) (Karaj, Iran) and Experimental Station near Borazjan (Bushehr Province of Iran) in 1999 (spring-summer season) and 2000 (winter season).

Released and experimental sunflower hybrids and lines of VNIIMK breeding were used as a material. To create segregating populations 2–3 elite lines were crossed with each other. Five Iranian fertility restorer lines (Rf-lines) (R-43, R-14, R-68, R-82 and R-217) were pollinated by five F₁ VNIIMK Rf-cross combinations (VK-580 × VK-571, VK-591 × VK-539, VK-700 × VK-580, VK-591 × VK-700 and VK-539 × VK-571). All of the obtained material along with commercial hybrids was self-pollinated. To obtain new initial material for B-line (maintainers of sterility) development four Iranian lines (CMS-31B, CMS-14B, CMS-19B and CMS-60/52B) were pollinated by four F₁ B-line cross-combinations from VNIIMK (VK-623 × VK-616, VK-680 × VK-616, VK-623 × VK-680, VK-680 × VK-456). All crosses were made without emasculation because inbred lines were used as a female parent and identification of hybrids in the offspring was not a problem.

Joint work of All-Russia Research Institute of Oil Crops (Krasnodar, Russia) and Seed and Plant Improvement Institute (Karaj, Iran) for elite line-breeding started in 2009 could be an example for sunflower shuttle breeding. During the spring-summer season, we obtained seeds of 76 F_1 and F_2 cross combinations in Karaj. Among them 40 populations for Rf-line development (25 F_1 and 15 F_2) and 36 populations for B-line development (15 F_1 and 21 F_2). All the material was planted in the South of Iran (Experimental Station near Borazjan in Bushehr Province) for self-pollination.

Best plants for all morphological traits were individually isolated for self-pollination in Borazjan. Low temperature during flower time in January creates comfortable conditions for fungi diseases, especially in isolated sunflower heads. Plants with symptoms of any diseases were discarded. Seeds of 35 F_1 and 33 F_2 cross combinations were obtained during the autumn-winter season. All the seed lots were divided into two equal parts – for use in Iranian and Russian breeding program separately.

The experimental design was randomized blocks with three replications. Each replication had four rows and two central rows were analyzed only to exclude the border effect. Oil content was evaluated by an NMR analyzer. Schneiter and Miller's method (1981) was used for the description of sunflower growth stages.

Results and discussion

Line breeding and evaluation take a lot of time (5–7 years) especially if you could not use off-season nurseries. After that, it is necessary to find a good combination, put a hybrid in a trial (up to 5 years in a breeding center), then in a State trial (2–3 years). The actual evaluation of a hybrid will come from the farms only. It is the reason why any breeder could make a reasonable decision about the effectiveness of his methods and strategies after quite a long period only.

Generally, the experiment for setting up the off-season sunflower nursery in the South of Iran was successful. However unusual cultivation conditions (short daylight period and low temperatures especially) cause some changes in plant growth and development. To evaluate the changes VNIIMK hybrid Kubanskiy 931 was used in all the locations in Iran as a check (Table 1).

Location	Days from emergency till the stage			Plant height, sm	Head diameter, sm
	R-1	R-5.1	R-9		
Karaj	30	51	91	147.0	18.0
Borazjan	37	57	107	115.6	10.8

Table 1: Characteristics of hybrid Kubanskiy 931 in different locations in Iran.

R-1, star stage (flowering bud became visual); R-5.1, beginning of flowering; R-9, Physiological maturity (Stage identification was made by Schneiter and Miller's method [1981]).

It is clear from the table that quantitative traits (plant height and head diameter) were decreased greatly, so this location could be used to obtain extra generation per year, but it was a stressful environment for the sunflower.

Starting from 2000 Russian part of obtained initial material was cultivated under usual for the sunflower environment at VNIIMK (Krasnodar). We used the ordinary pedigree technique for sunflower line development (Kaya et al. 2012; Vear 2010). Breeding for resistance to broomrape and diseases is very significant for our region (Gontcharov 2009; 2014). Lines, produced at the end of the process, were much more resistant to abiotic stresses than usual.

We consider it as an obvious effect of the shuttle breeding technique. Individual plant selection was made in very different climatic, soil and cultivation conditions:

- 1) Karaj location mountain desert with high differences between day and night temperatures, very dry air and irrigation once in 3–5 days by channels;
- 2) Borazjan location wintertime, relatively low temperature during flowering time, the short period of daylight, the same type of irrigation as in the previous location;
- 3) Krasnodar location a long period of daylight, high temperature during flowering, deep fertile soil, no irrigation. As a result, we get a set of lines with an unusually high percentage of prospect ones.

General results are shown in Table 2.

Seventy-six initial populations from Borazjan were cultivated in Krasnodar, Russia. Several initial material populations were discarded, and only 24 left. Among them we get 43 elite lines, many of them became parental forms of released and prospect hybrids. Characteristics of several inbred lines are presented in Table 3.

Type of cross	Borazjan,	Krasnodar		
	1999–2000	Population number, 2009	Population 2009, %	Elite lines number, 2019
F ₁ for B-line development	15	1	6.7	0
F ₁ for Rf-line development	25	5	20.0	7
F ₂ for B-line development	21	13	61.9	29
F ₂ for Rf-line development	15	6	40.0	7
Total F1	40	5	15.0	7
Total F2	36	19	52.8	36

Table 2: Number of initial populations and developed sunflower lines.

Line	Туре	Origin	Period from emergency to flowering, day	Plant height, sm	Bonus traits*
VK-905	В	VK-680 × (VK-639 × VK-464)	48	132	HO, EM, LS, OR-5, GCA
VK-934	В	VK-680 × (VK-639 × VK-464)	52	145	HO, LS, OR-5, GCA, PhT
VK-900	В	VK-680 × (VK-653 × VK-464)	54	150	HO, OR-5, GCA, PhT
VK-930	Rf	(VK-678 A × VK-653) × VK-580	50	143	DM (330), PhT, GCA
VK-920	Rf	R-217 × (VK-580 × VK-571)	48	120	DM (330), EM, PhT, GCA
VK-915	Rf	(VK-580 × VK-571)	46	128	DM (330), EM, PhT, GCA

Table 3: Characteristics of the some elite sunflower inbred lines, developed by the method.

*Notes: HO, high oleic, EM, early maturing; LS, large seeds, suitable for confectionary hybrid production; OR-5, resistant to the race E of broomrape; GSA, high general combining ability; PhT, high tolerance to Phomopsis; DM (330), resistant to downy mildew (race 330).

All developed sunflower inbred lines have high oil content, very high adaptability to different environments, high general combining ability, good resistance or high tolerance to the most dangerous diseases. All this traits made them suitable for different breeding programs. They are used as parental lines in a set of prospect and released hybrids.

Conclusions

Comparing F_1 and F_2 population results we concluded that the application of shuttle breeding technique to sunflower was highly successful and results in elite line development. The low percentage of lines developed from F_1 populations have stressed the effectiveness of the shuttle breeding method because any selection from the F_1 population has no sense and all the next generations were cultivated in Krasnodar only. It is shown that the application of the shuttle breeding technique to sunflower was highly successful and results in elite line development.

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