

HYBRIDIZATION BETWEEN SUNFLOWERS (*HELIANTHUS ANNUUS* L.) AND LESS STEM ROSETTE (*CARLINA ACANTHIFOLIA* ALL.). CHARACTERIZATION OF RECEIVED INTERGENERIC FORMS

Mihail Christov¹, Miroslava Hristova-Cherbadzhi²

¹Mihsan Ltd, Sofia, Bulgaria; ²University of Forestry, Sofia, Bulgaria

ABSTRACT

Hybridization between sunflower (*Helianthus annuus* L.) and less stem rosette (*Carlina acanthifolia* All.) were made. These were two plants with different habitat in northern Bulgaria. Intergeneric hybrid plants and seeds were obtained in both directions of crossing. There were some outstanding differences between the two intergeneric hybrid groups. The first is related with the pollination and the viability of hybrid plants. Another interesting difference was in the seeds. Seeds of second group plants were larger, with 2-3 times longer size than those from first group, though the maternal parent form was the same. Line had small seeds. New intergeneric hybrids were carriers of *Rf* genes for CMS Pet-1 transferred from *Carlina acanthifolia* All. In crosses with sterile sunflower lines showed 100 % restoration ability.

Key words: *Carlina acanthifolia*, *Helianthus annuus*, intergeneric hybrid, sunflower

INTRODUCTION

Application of intergeneric hybridization in sunflower is hardly realizable work. Under certain conditions and well-chosen parental components enables the creation of rich source material for selection of sunflower (Christov and Panajotov, 1991; Christov et al., 1994, 2004, 2009; Christov and Vassilevska-Ivanova, 1999; Hristova-Cherbadzi, 2007, 2012; Christov, 2013 and etc.). Of particular interest are the plants used in folk medicine. One of these plants is less stem rosette (*Carlina acanthifolia* All.).

MATERIAL AND METHODS

The investigation was carried out at the Vrachantsi, Dobrich, Bulgaria, during the period 2013 - 2015.

For maternal parent form used sterile analogue of line HA-821 and as pollinator *Carlina acanthifolia*: accession №1, found near Vrachantsi, Dobrich (Fig.1) and accession №2 by region Balgarevo, Kavarna.



Fig.1. *Carlina acanthifolia*.

Method of intergeneric hybridization (crossing between cultivated sunflower line HA-821A and *Carlina acanthifolia*), crossing between A lines and intergeneric hybrids, selection,

self-pollination and sib-pollination were used. Morphological characteristics were based on phenotypic observation and biometric measurements during the vegetation period and on laboratory studies of whole plants and seeds. *Rf* genes, in crosses of sterile sunflower lines x *Carlina acanthifolia* and sterile sunflower lines x intergeneric hybrid - F₁ and F₂, were searched. The taste of the kernel is tested in wax and full maturity and after short roasting seeds.

RESULTS AND DISCUSSION

As a result of hybridization between sunflower (*H. annuus*) - line HA-821A and *Carlina acanthifolia* - accessions №1 and №2 total 31 seeds and from them 7 F₁ plants were obtained (Table 1). The number of received seeds for separate inflorescences was from 1 to 8.

Table 1. Crossability of cultivated sunflower *H. annuus* and *Carlina acanthifolia*.

Crosses	Pollinated inflorescences			Total number seeds	Hybrid plants	
	total number	with seed			number	%
		number	%			
<i>H. annuus</i> x <i>C. acanthifolia</i> №2 - “first group”	23	5	21,74	12	2	16,67
<i>H. annuus</i> x <i>C. acanthifolia</i> №1 - “second group”	23	3	13,04	19	5	26,32

In early March 2014 the seeds were sown in separate containers in a special room with enough light and warm (21-25⁰C). Then they were seedlings in 30 cm distance from place, where was the *C. acanthifolia* accessions №1. The plants developed normally until flowering. Their height was from 30 to 55 cm. The disk flowers of all plants digressed pollen. One plant from the “second group” (*H. annuus* x *C. acanthifolia* №1) died 3 days after full of blooms. Another plant from the “first group” (*H. annuus* x *C. acanthifolia* №2) died 21 days after blooming. After self-pollination of the plant from the “first group” and two plants from the “second group” are received in total 11 (4 + 7) seeds. Two plants from the “second group” were pollinated with one another. After that 17 seeds were obtained. With pollen from the last two plants was pollinated one sterile plant from the line 92A. After this bekkros number of seeds was 21. In 2014, a second generation plants (2 and 1) were received also. From each group are left spare seeds. Results of the seeds' viability from the first hybrid generation and the viability of hybrid plants from the next generation are presented in Table 2.

Table 2. Viability of seeds and hybrid plants from the second generation, 2014.

Type of pollination	Sown seeds, n	Received plants, n	Vital plants	
			number	%
Self-pollination				
- “first group”	3	2	2	100,00
- “second group”	5	4	4	100,00
Sisterly pollination				
- III group	12	11	10	90,91
Bekkros				
- IV group	15	15	15	100,00

From sown total 8 seeds after self-pollination of F₁ plants, were received 6 F₂ plants. The disk flowers of all plants digressed pollen. After self-pollination of the two plants from “first group” 32 seeds were obtained. Their dimensions were closed to these of line HA-821A. Some of the seeds had gray-black color of the peel and the others were light- motley colored. After self-pollination of 4 F₂ plants from the “second group”, 89 seeds were received. The seeds are characterized by large size in length (12-13 mm) and light- motley colored. From III group (since the sisterly pollination of the F₁ plants) 12 seeds were sown. Eleven F₂ plants were obtained. At the beginning of flowering one plant dies from them. The other 10 F₂ plants were fertile. Seven from them were self-pollinated and 201 seeds were obtained. The remaining three plants were left open pollinate. Since all plants the seeds were obtained, but at about 1/3 of the disc florets on each head had not seeds. The best result was obtained at BC₁ plants from the IV group.

From 15 sown seeds, 15 vital plants were received. One from them was male sterile and the other 14 plants - fertile. Eight plants were self-pollinated and 7 (6 + 1) plants were left open pollinate. From all 15 plants, the seeds were obtained. By two inflorescences of lines HA-821A and 92A were pollinated with pollen from F₂ plants (cross *H. annuus* x *C. acanthifolia* №1, “second group”). Seeds were obtained from the 4 pollinated heads. Twenty one seeds from all the numbers of different groups’ intergeneric hybrids were sown on April 2015 to obtain plants from third generation. Some results are presented in Table 3.

Table 3. Characteristics of F₂, BC₁, F₃, F₁, BC₁ and F₄ hybrids.

Groups	Sown seeds, n	Received plants, n	Vital plants, n	Inflorescences with seeds, n	Male sterility plants, n
I group	21	17	17	9	-
II group	21	19	19	19	-
III group	21	16	16	16	1
IV group	21	20	20	20	1
V group:					
1 head - HA-821A x II group	21	20	20	20	3
2 head - HA-821A x II group	21	19	19	19	2
1 head 92A x II group	21	21	21	21	2
2 head - 92A x II group	21	20	20	20	1

Six plants from all numbers (groups) were self-pollinated, to receive next generation.

Some of the F₃ plants originating from a cross HA-821A x *C. acanthifolia* №2 had not seeds, although the plants were fertile. Seven of the 8 plants that have not received seeds were open pollinated. This may mean that the work with accession №2 will be difficult. Probably the two accessions №1 and №2 were different. The seeds from all plants II group are distinguished with their length (12-13 mm) and light-motley colored. The seeds from more of plants III group were similar to those of the II group. From other plants, seeds larger than the line HA-821A were received. All plants from the IV group have large seeds, because line 92A was with large seeds. Some of the seeds are colored gray-brown. The seeds of the first two numbers from the V group were larger than those of the line HA-821A, but were smaller than those of the plants from the II group. Seeds from the other two numbers were long and similar to the seeds of plants from the II group. The received fertile plants from all groups showed that in both accessions were established *Rf* genes for CMS Pet-1.



The taste of the kernel from the F₃ plants II group is tested in wax and full maturity and after short roasting seeds. As standards are used variety Favorit and hybrid XL-4337. Taste of nut plants from II group compared with this from nuts on both standard was more similar to hybrid XL-4337, but different in wax maturity.

CONCLUSION

Hybridization between sunflower (*H. annuus*) and two accessions of *C. acanthifolia* was successful. The received interspecies hybrids with the both accessions were differ in some characteristics, such as size of the seeds, in the plant growth and the next generation multiply, carriers of *Rf* genes for CMS Pet-1 and others. Many of the characteristics of the received material to be studied for the future.

ACKNOWLEDGEMENTS

We would like to thank *Mihsan Ltd*, Bulgaria that gave us the financial support to work, which led to the successful production of the new materials described in this paper.

LITERATURE

Christov, M., 2013. Contribution of interspecific and intergeneric hybridization to sunflower breeding. *Helia* 36(58): 1-18.

Christov, M. and Panajotov, I., 1991. Hybrids between the Genera *Helianthus* and *Tithonia* and their study. *Helia* 14(15): 27-34.

Christov, M. and Vassilevska-Ivanova, R.D., 1999. Intergeneric hybrids in *Compositae* (*Asteraceae*). I. Hybridization between cultivated sunflower *H. annuus* L. and *Compositae* Genera. *Helia* 22(31): 13-22.

Christov, M., Hristova-Cherbadzhi, M., Nikolova, V., Ivanova, I., Shindrova, P., 2009. Intergeneric hybridization to sunflower – results and problems. In: Proceeding of international

scientific conference „Good practices in sustainable agriculture”, Sofia, 12-13 November 2009, 167-184.

Christov, M., Kiryakov, I., Shindrova, P., Encheva, V., and Christova, M., 2004. Evaluation of new interspecific and intergeneric sunflower hybrids for resistance to *Sclerotinia sclerotiorum* (Lib.) de Bary. Proc. 16th Int. Sunflower Conf., Fargo, North Dakota, USA, August 29-September 2, 2004, vol. II, 693-698.

Christov, M., Vasileva, R., Tsujimoto, H. and Panajotov, I., 1994. Intergeneric hybridization between sunflower and some species of genera from *Compositae*. International *Compositae* Conference, Royal Botanic Gardens, Kew, 26.07-05.08, p. 80.

Hristova-Cherbadzhi, M.M., 2012. Study of new forms of sunflower received by distant hybridization. Breeding and genetics of cultivated sunflower - methods, new lines, new crosses, new cms source. Lambert Academic Publishing, ISBN 978-3-659-13617-7.

Hristova-Cherbadzi, M., 2007. Study of new forms of sunflower received by distant hybridization. PhD thesis, BAN, Sofia.