Review Article

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The top Bulgarian contributions to sunflower breeding

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Abstract: The sunflower (*Helianthus annuus* L.) is a fundamental oil-yielding culture for Bulgaria and the world and thus, special attention is paid to it. Various selection methods have been used over the years and as a result of successful breeding programs new sunflower forms, lines (*B*/*A* and *R*) and hybrids (oil and confectionery type) were created in Bulgaria. Some of these new forms have new plant architectonic, different vegetation periods, seeds with different sizes and coloration, high combining ability, resistance to diseases, the parasite Broomrape and some herbicides, high seed oil and fatty acid content, genes for restoration of fertility (*Rf* genes), and different types of cytoplasmic male sterility (*CMS*).

Keywords: achievements; *Helianthus*; heterosis selection; hybridization; sunflower breeding.

1 Introduction

"*Helianthus annuus* L. is the best-known sunflower across the world. This "common sunflower" is extremely variable. It has many different forms – branched (they have small flower heads and are common in the wild, mainly western North America); unbranched (with massive flower heads and cultivated for their oily seeds); and other forms (which are grown for their ornamental value expressed by their red or double flowers)" (Heiser 1981). According to the latest classification, the genus *Helianthus* has 51 species, 14 of which are annual and 37 – perennial (Seiler and Gulya 2004).

Thanks to archaeological evidence from North America – as all wild species of the genus Helianthus originate from there (Heiser 1978), it has been revealed that species of the genus have been used from as far ago as around 3000 BC

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(by Native Americans) (Heiser 1945). Although the genus was introduced much later to Europe, as the first sunflower seeds were sown in 1510 in the Madrid Botanical Garden (Zhukovsky 1950), it quickly spread – by the end of the 16th century, from Spain to most Western European countries (Zimmermann 1958), later on, in the 18th century, to Russia (Gundaev 1971) and nowadays, all over the world – in about 80 countries (https://www.atlasbig.com). That is not surprising since the cultivated sunflower (*Helianthus annuus* L.) has a wide range of distribution – from 30° to 50° north latitude and from 10° to 40° south latitude (Škorić et al. 1989) and since all parts of the sunflower plant are of economic importance (Škorić 1988). Additionally, *Helianthus annuus* L. is one of the five main sources of vegetable oil in the world and some of the best quality edible oils are obtained from it (Škorić 1988). This specie has many more applications – it is used in the medicine, textile, paper, cosmetic, and soap industries, and as a biofuel (oleic type). The sunflower's economic importance can be easily observed by its success in the international market. The introduction of *Helianthus annuus* L. as an oil crop in Bulgaria took place during the First World War (Petrov 1962). After 1927, the first Bulgarian varieties No. 3–18 and No. 15–16 were created and implemented in production (Petrov 1962). Since then a lot of changes have occurred most notably, the sunflower selection in Bulgaria has gone through several stages:

(1) local production of materials from other countries;
(2) obtaining new varieties through the use of intraspecific (interline and intervarietal) and distant (interspecific and intergeneric) hybridization and selection;
(3) using heterosis selection to create new hybrids.

At present, for Bulgaria, the sunflower is the second most important agricultural crop after wheat and the main oilseed crop (MZH 2021). From 2011 to 2020, between 747,100 ha (2011) and 898,844 ha (2017) were sown with sunflower, and the average yield varied from 1.78 t/ha (2012) to 2.44 t/ha (2018) (MZHG). In addition, in the period 2018–2020, Bulgaria was the 6th largest producer of sunflowers in the world by obtaining tons and the 7th by sown hectares (https://www.atlasbig.com).

Various selection methods have been used over the years to obtain new Bulgarian varieties – classical ones such as intraspecific and remote hybridization (Bohorova 1977; Bohorova and Georgieva-Todorova 1987; Cvetkova 1976, etc.) and experimental mutagenesis (Christov 1996a), as well as new methods such as the biotechnological (Bohorova and Atanassov 1990; Nenova et al. 1998; Punia et al. 1994; Todorova et al. 1997a, b; Yordanov et al. 2005) and molecular ones (Spassova et al. 1992a, b). The use of different methods aims to increase genetic diversity by taking into account the changes in the plant yields, resistance to diseases, morphological structure as well as the quantity and quality of the oil in the seed.

Some of the best results of Bulgarian sunflower breeding are presented in this article.

2 Materials and methods

The included **materials** are different varieties and lines, sterile analogues of cultivated sunflower *Helianthus annuus* (2*n* = 34), wild species from the genus *Helianthus* and other genera of family *Compositae,* as well as numerous hybrid materials.

2.1 Introduction of species

The first step in the creation of new varieties in Bulgaria was to set up a collection of wild species. It happened 40 years ago at the Dobroudja Agricultural Institute (DAI), General Toshevo (Figure 1). 40 *Helianthus* species with different ploidy levels – 10 annuals and 30 perennials (Figures 2 and 3), as well as 30 species from other genera of family *Compositae* (Figure 4) made up this collection (Christov 1996b; Hristova-Cherbadzhi 2012, etc.). Many foreign scientists visited it which contributed to the exchange of materials between the members of the established FAO groups. After that, the results were jointly discussed at conferences (Figure 5).

Figure 1: Collection of wild species in the Dobroudja Agricultural Institute, General Toshevo.

Figure 2: Wild *Helianthus* species.

Methods of hybridization and selection were used to obtain the results presented below. Self-pollination, sib-pollination, crossing and backcrossing were implemented. The pollen from the cultivated sunflower and that from different interspecific and intergeneric hybrids were used (Figure 6). Evaluation for resistance to diseases, the parasite broomrape and herbicides were also carried out (Christov et al. 1992; Christov 1996c; Christov et al. 2004, etc.). Furthermore, seed oil was analyzed. On the base of phenological observations and biometric measurements during the vegetation period, a morphological characteristic was made.

In the past, combinatorial selection whose task is the creation of a new, stable (direct) variety was used. With time, however, it was observed that when hybrids were sowed the yield increased (Putt 1997) and as a result, today the focus has shifted to heterosis selection. This type of breeding aims to create and select suitable lines of parent pairs, from the crossing of which a heterosis hybrid offspring that can be used directly for production purposes is obtained. To have a strong heterosis effect, the pairs of homozygous parental lines should have high general and specific combinatorial abilities. Only the first generation (F1) is used because in the subsequent generations the heterosis effect decreases due to genetic breakdown.

CMS lines (A) – sterile analogues of fertile (B) lines in which the qualities of the fertile lines are preserved, are used in hybrid seed production. This saves time by avoiding the long and labour-

Figure 3: *Helianthus* species: 1. Annual: a – *H. argophyllus* (E-007), b – *H. praecox* ssp *hirtus* (E-027), c – *H. bolanderi* (E-009), d – *H. neglectus* (E-017), 2. Perenual: e – *H. pumilus* (M-172), f – *H. nuttallii* ssp. *rydbergii* (GT M-173/№18), g – *H. maximiliani*, h – *H. microcephalus*, i – *H. mollis*, j – *H. pauciflorus* ssp. *subrhomboideus* (M-002), k – *H. xlaetiflorus* (M–005), l – *H. maximiliani*, m – *H. hirsutus* (GT M-007).

intensive castration of the more complex inflorescences and guarantees the purity of the maternal parent from its pollen. The fertile F_1 hybrid plants are obtained from the seeds that are produced after crossing the two parental forms – A and R lines. The *CMS* system for hybrid seed production has been used in *Helianthus annuus* since 1972 (Fick and Miller 1997).

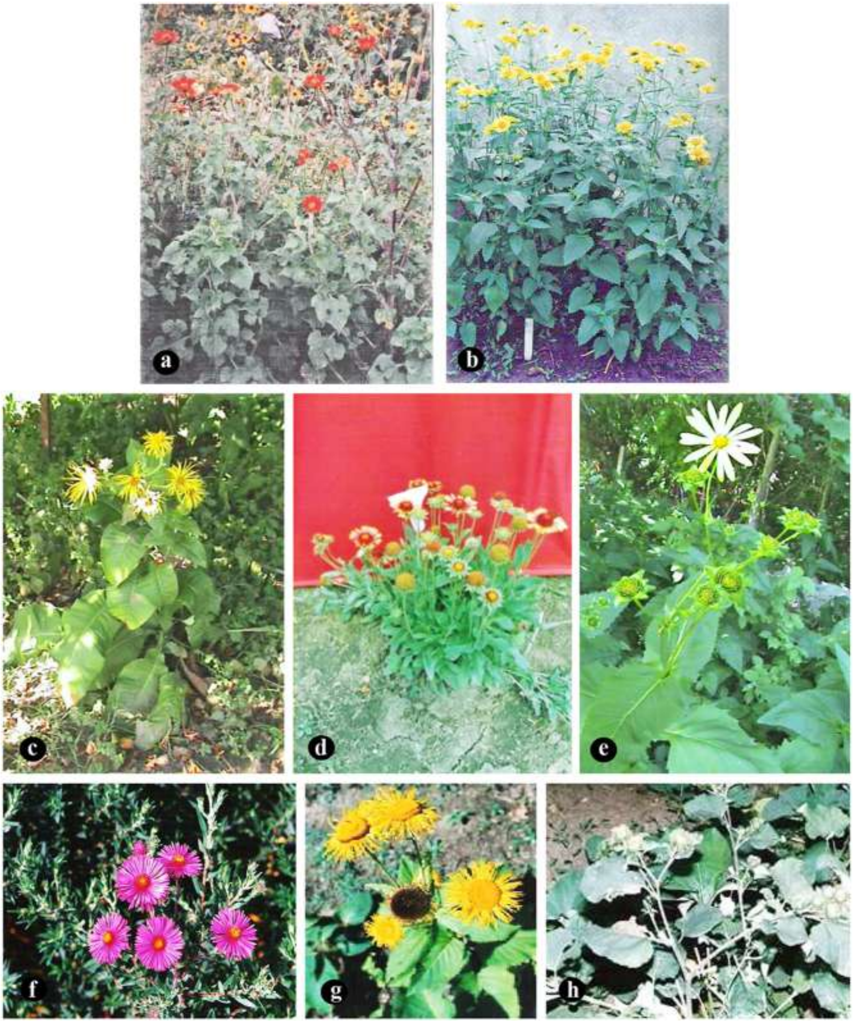

Figure 4: Species from genera of family Compositae: a – *Tithonia speciosa* ($2n = 34$), b – *Verbesina encelioides* (annual), c – *Inula helenium* (White oman, “бял оман”, $2n = 20$), d – *Gaillardia aristata* ($2n = 22$), e – *Silphium perfoliatum* ($2n = 24$), f – *Aster speciosa* ($2n = 72$), g – *Telekia speciosa* (Yellow oxeye “chernokos”), h – *Arctium lapa* (Lappa, “репей”, $2n = 32$).

3 Results and discussion

Bulgarian breeding has made a great contribution to the creation of new and diverse genetic materials that have been used all over the world. Chronologically speaking, the country’s first recognized achievement was the new Bulgarian broomrape-resistant sunflower variety “Tolbukhin 75” (registered in 1948). Later on, the hybrid

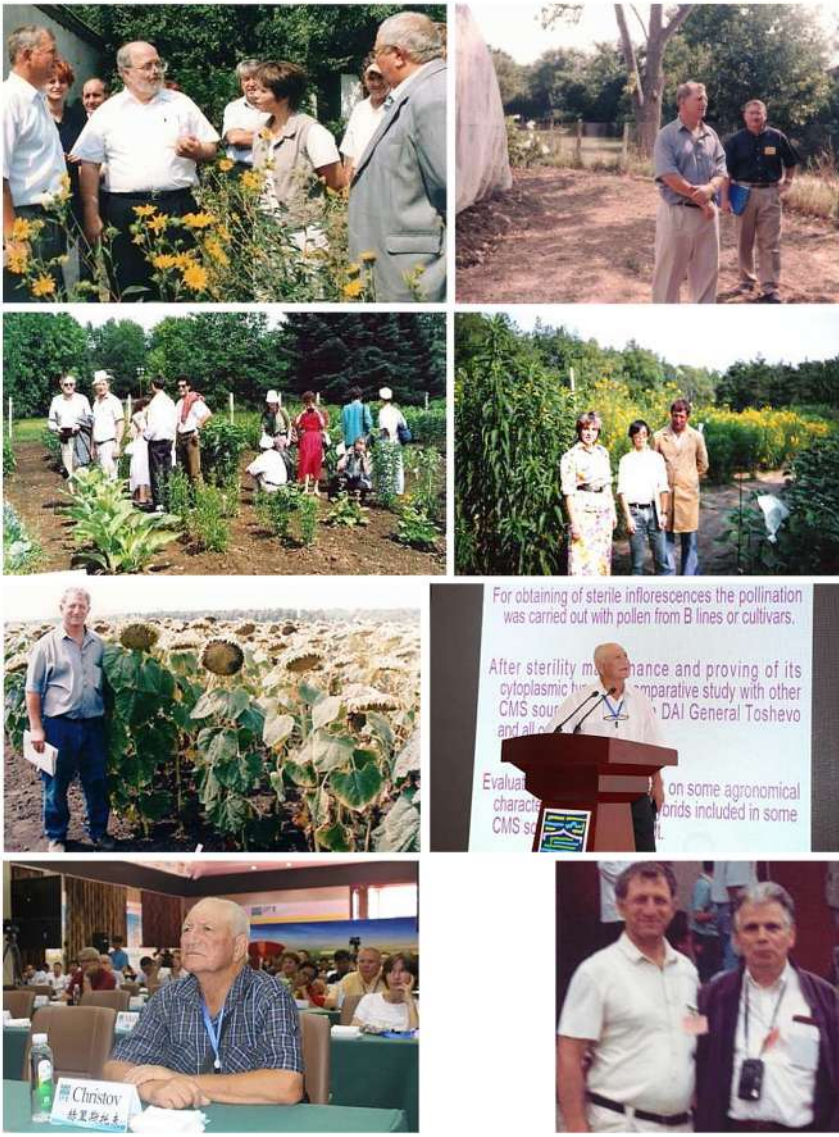

Figure 5: Material exchange, joint discussions in FAO groups and at conferences.

“Albena” (recognized in 1998) which is from the group of early-maturing hybrids played a key role in worldwide sunflower production. In fact, according to the unofficially disclosed information from “Prograin *Génétique*” at one point “Albena” occupied about 80 % of the sunflower fields in Europe.

Figure 6: Cross-pollination.

Until 2009, 8 varieties and 34 hybrids of Bulgarian origin were accepted and entered in the official variety list by the Executive Agency for Variety Testing, Field Inspection and Seed Control – EAVTFISC (Hristova-Cherbadzhi 2009a). As of 2022, the number of Bulgarian varieties approved for seed production has increased to 60 (EAVTFISC). Some of the best hybrids obtained over the years are Maritsa, Musala, Mesta, Mura, Madan (the last one being confectionery – suitable for human consumption) and others. The hybrid "Maritsa" (2004), which is on the official list of Russia, Ukraine and Moldova and is resistant to broomrape, was created after crossing lines 197 A and 7009 R. The paternal form (the R line) was obtained after self-pollination and the selection of an interspecific hybrid (in the making of which the wild species *H. tuberosus* M-037 participated) (Figure 7). The hybrid Musala,

Figure 7: Hibrids Musala and Marica, and line 7009 R.

which has the formula 2607A × 7043R (*H. pauciflorus* M-028 is involved in the paternal line), is resistant to *phomopsis* and tolerant to *sclerotinia*.k

After years of test and error, not only intraspecific heterosis hybrids from cultivated sunflowers were created but also interspecific and intergeneric ones from *H. annuus* with *species* from the genus *Helianthus* and other genera of family *Compositae* (Christov 1988a, b, 1991, 1996b; Christov and Hristova-Cherbadzhi 2016; Christov and Panayotov 1991; Christov and Vassilevska-Ivanova 1999; Hristova-Cherbadzhi 2009b; Hristova-Cherbadzi and Christov 2008a, b; Hristova-Cherbadzi et al. 2008; Nikolova et al. 1995, 2004; Nikolova and Christov 1996, 2004a; Valkova et al. 2008; Hristova-Cherbadzhi 2020, etc.) (Figures 8 and 9).

From zero to one hundred:

1. The process of interspecific hybrid making

The distinguished Bulgarian interspecific hybrids were obtained with the participation of species from all ploidy groups of the genus *Helianthus* in one (straight cross) or both directions of crossing (reciprocal crosses). In the process of making the following correlations were discovered: the percentage of hybrid plants that were developed was higher when the cultivated sunflower was used as the maternal parent; the percentage was also influenced by the wild species included in the cross; crossing with the annual diploid species was more effective (to varying degrees) than with the perennial diploid species, regardless of the equal chromosome number in their genomes; the cultivated sunflower was the most easily crossed with perennial species with a hexaploid set of chromosomes, then with tetraploids and with the most difficulty with diploid species. An additional noteworthy observation is that the inheritance in the hybrids of the first generation was intermediate or closer to the wild type – most of the F1 hybrids have a one-year growth cycle (when all the perennial diploid species and other species are involved) and others are multi-year growth cycle (when some of the perennial hexaploid and tetraploid species are included). In all cases, however, the hybrid plants of the next generations from the direct and reciprocal crossing have a one-year development cycle.

There are many more factors that affect the degree of hybridization and zygote production. To name a few – the time of hybridization, the vitality and amount of used pollen, the direction of crossing, the time divergence during the flowering of the perennial species of the genus Helianthus with the cultivated sunflower, climatic conditions, etc. More specifically, in dry and hot weather fertilization is weak, the seed set is moderate, and the germination of the derived seeds is low. Besides that, using fresh pollen increases the number of obtained seeds.

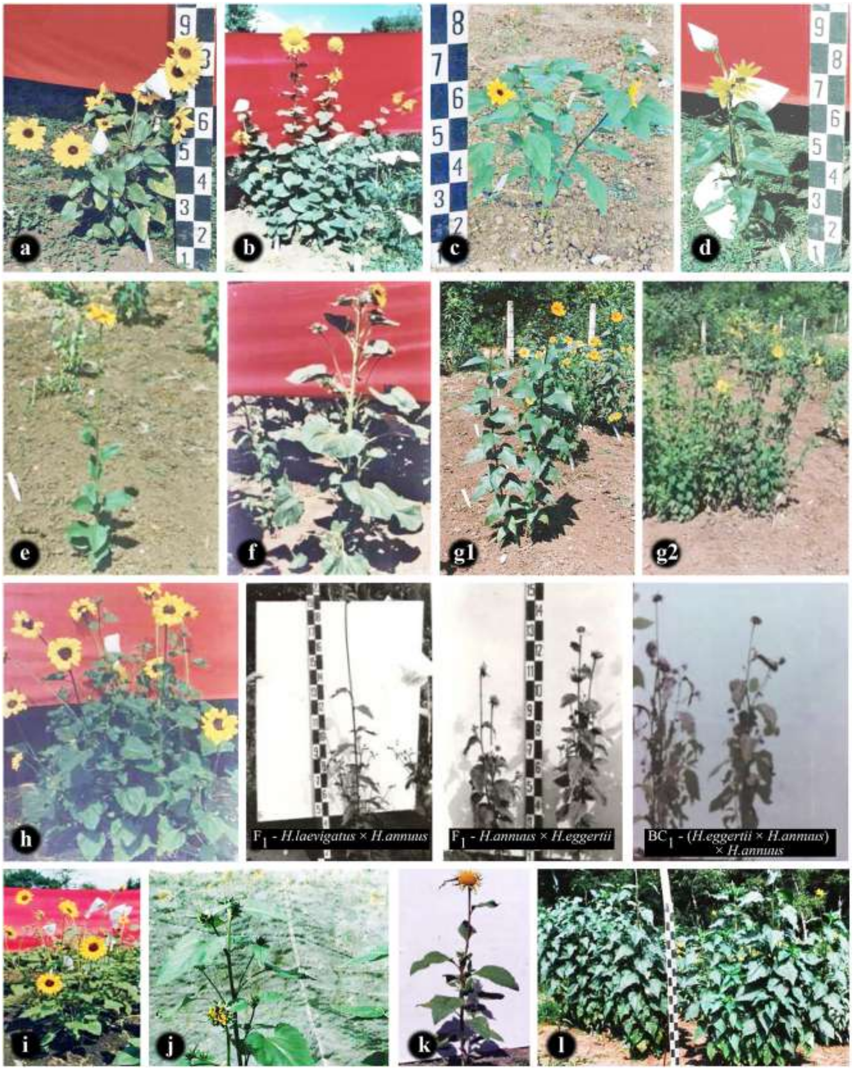

Figure 8: F1 interspecific hybrids: a – *H. annuus* × *H. bolanderi*, b – *H.praecox* ssp *hirtus* × 1234B, c – *H. annuus* × *H. giganteus*, d – *H. annuus* × *H. nuttallii* ssp. *rydbergii*, e – *H. annuus* × *H. x. laetiflorus*, f – *H. annuus* × *H. strumosus*, g – 1. *H. annuus* × *H. hirsutus*, and perennial F1 – 2. *H. hirsutus* × *H. annuus*, h – *H. annuus* × *H. mollis*, i – *H. annuus* × *H. debilis*, j – (*H. hirsutus* × *H. annuus*) × *H. annuus*, k – BC1F1BC1 with *H. decapetalus*, l – *H. annuus* × *H. tuberosus*, and etc.

2. New lines characterized by high combining ability

The Bulgarian inbred lines are characterized by high combining ability which makes them highly sought. The height of the plants varies from 0.90 to 1.80 m, and the diameter of the inflorescence – from 13 to 27 cm. The number of seeds per inflorescence is from 800 to 2000 for the unbranched and from 650 to 1300 for the

Figure 9: Intergeneric hybrids: a – *H. annuus* × *Bidens tripartite*, b – *H. annuus* × *Verbesina helianthoides*, c – 1. F2 plants from cross *H. annuus* × *Matricaria chamomilla*, 2. B line and 3. R line, d – seeds from parents F1 cross *H. annuus* × *Tithonia rotundifolia*.

branched. The yield of seed (in weight) from one inflorescence for unbranched plants is from 48 to 110 g, and for branched plants from 20 to 70 g. For the R lines, the mass of 1000 seeds varies from 20 to 65 g, and for the B lines mostly around and above 60 g. The vegetation period is from 100 to 140 days. While the new B lines are unbranched, the majority of R lines are branched (Table 1).

Table 1: Characterization of some lines.

No	Origin	Plant height (cm)	Head diameter (cm)	Vegetation period (days)	Seed oil content (%)	Resistance or tolerance to
			R lines			
1018	*H. nuttallii* ssp. *rydbergii* M-172	140	16	106	53.4	downy mildew
1411	*C. acanthoides*	105	12	96	48.2	downy mildew
2499	*H. pauciflorus* ssp. *subrhomboideus* M-002	130	22	105	52.3[a]	downy mildew
2505	*H. nuttallii* ssp. *rydbergii* M-172	135	14	104	60.8	downy mildew/ phomopsis/ phoma/ broomrape
2536	*H. neglectus* E-017	125	13	107	49.2	downy mildew/ phomopsis/ broomrape
2879	*H. pauciflorus* ssp. *subrhomboideus* M-002	125	16	110	52.1	
2906	*H. pauciflorus* ssp. *subrhomboideus* M-002	120	21	103	52.1	downy mildew/ broomrape
3209	*H. neglectus* E-017	120	12	102	53.9	
3292	*H. hirsutus* M 029	120	21	108	50.6[a]	downy mildew/ phomopsis/ sclerotinia/ broomrape
7004	*H. praecox* E-028	145	16	112	52.6	
7009	*H. tuberosus* M-037	80	13	92	46.0	broomrape
7015	*H. debilis* E-011	120	15	102	50.7	
7009	*H. tuberosus* M-037	80	13	92	46.0	broomrape
7026	*H. smithii* M-008	140	14	106	45.4	broomrape
7041	*H. eggertii* M-001	120	15	101	47.2	phomopsis
7043	*H. pauciflorus* M-028	135	16	106	52.5	phomopsis/ sclerotinia
7070	*C. acanthoides*	130	14	98	48.4	sclerotinia
7082	*H. glaucophylus* M 012	110	16	107	49.9	
7083	*T. rotundifolia*	90	13	97	50.1	
7090	*H. paradoxus* E-019	120	15	104	52.1	
7091	*H. ciliaris* M-092	70	21	105	45.3	
7092	*H. divaricatus* M-044	120	17	105	52.4	sclerotinia
C 55	*H. debilis* E-011	120	16	105	52.7	broomrape

Table 1: (continued)

No	Origin	Plant height (cm)	Head diameter (cm)	Vegetation period (days)	Seed oil content (%)	Resistance or tolerance to
PR 35/5	*H. divaricatus* M-044	125	16	109	51.7	
PR 50/5	*H. neglectus* E-017	120	16	109	48.6	downy mildew/ phomopsis
			B lines			
1305	*H. debilis* E-014	150	21	108	52.7	broomrape
2942	*H. pauciflorus* ssp. *subrhomboideus* M-002	140	24	116	52.5	downy mildew/ broomrape
3291	*H. hirsutus* M 029	110	23	118	50.6	downy mildew/ phomopsis
6101	*H. decapetalus* M-043	125	18	106	47.4	downy mildew/ broomrape
6134	*H. debilis* E-011	100	22	107	48.1	downy mildew/ broomrape
6149	*H. eggertii* M-001	140	24	103	48.9	downy mildew/ broomrape
6191	*H. decapetalus* M–043	150	18	109	52.5	downy mildew
6215	*H. salicifolius* M–045	180	18	107	51.2	
6217	*H.* × *laetiflorus* M-005	120	17	110	49.7	
6275	*H. argophyllus* E-007	140	23	105	50.0	downy mildew/ broomrape
6291	*H. debilis* E-011	155	24	108	47.1	downy mildew/ broomrape
6330	*H. praecox* E-029	125	16	113	45.4	
6748	*H. decapetalus* M-043	118	19	108	45.9	phomopsis
M-8-A1	*H. neglectus* E-017	130	21	113	49.32	downy mildew

[a]unbranched plants.

Over the years, as a result of working with hybrid offspring, B/A and R lines have been created with valuable economic qualities such as:

- complex resistance to *Plasmopara helianthi*, *Phomopsis helianthi*, *Phoma macdonaldii*, *Alternaria helianthi*, and *Orobanche cumana* and high tolerance to *Sclerotinia sclerotiorum* (Batchvarova et al. 2001; Christov et al. 1998, 2009) (Figure 10);
- resistance to herbicides (Christov et al. 2008; Christov and Hristova-Cherbadzhi 2020; Hristova-Cherbadzhi 2022);

– with high oil content in the seeds (53–60 % – with the participation of *H. nuttallii*, 52 % – with *H. pauciflorus*, 51 % – with *H. hirsutus*, etc.) and varied fatty acid composition of the oil and amino acid composition of the protein (Hristova-Cherbadzhi 2012);
– seeds with different sizes and colours (Figure 11).

Some of the lines were also characterized by molecular markers (Atanasova et al. 2004; Hvarleva et al. 2007, 2009) and by statistical methods (Hristova-Cherbadzhi and Molle 2016).

The carried out remote hybridization allows diverse hybrids and valuable starting material for the selection of sunflowers. Currently, some of these lines are certified and can be used practically in seed production for the creation of new heterosis hybrids (Figure 12).

The development of *B* lines – ***Maintainer of sterility,*** is done by targeted selection and self-pollination in the hybrid materials, which begins in the second or third generation and can continue up to the 13th one. The evaluation of materials is based on their morphological, biochemical and phytopathological characteristics, as well as the absence of *Rf* genes and the presence of good combining ability.

After purposeful and repeated selection and self-pollination ***R* lines** – ***Restorer of fertility*** can be created to homogenize (equalize) and bring *Rf* genes into a homozygous state. Forms with *Rf* genes can also be examined in materials obtained from crosses of *cultivated sunflower* (*B* line) × *wild species* or *wild species* × *cultivated sunflower*. The development of these forms is done by pollination of sterile plants from a cultivated sunflower based on a *CMS* source. Some *R* lines which possess 100 % fertility restoration in addition to other important characteristics were obtained in Bulgaria (Figures 13 and 14) (Christov 2008, 2013; Hristova-Cherbadzi and Christov 2008c).

The discovery of *Rf* genes' souses is done by crossing *sterile sunflower line × wild species.* The sterile line can be included in different *CMS*. Because of this the newly discovered *Rf* genes may not only differ from each other but also may be present in varying qualities (Christov 1992c; Hristova-Cherbadzhi 2012; Nenov et al. 2002). After pollination of sterile analogues of lines *H. annuus* in CMS PET1 with pollen from wild species, whose genome does not carry *Rf* genes, all derived F_1 plants would be male sterile, as the maternal line is with genotype *rfrf*. If the species of genus *Helianthus* carry genes restorer of fertility (*Rf*) for CMS PET1 all or part of the received F_1 plants would be fertile. For the restoration of the fertility of CMS ARG3-M1, 2 *Rf* genes are necessary.

Discovery of *CMS* souses is possible in the cross of *wild specie × cultivated sunflowers*. If a sterile F1 plant is obtained, it must be crossed with an anchoring B line. After several consecutive sterile generations, which are obtained after pollination with the B line, it can be proven that the cytoplasm (basically the

Figure 10: Evaluation of resistance to *sclerotinia* and herbicides.

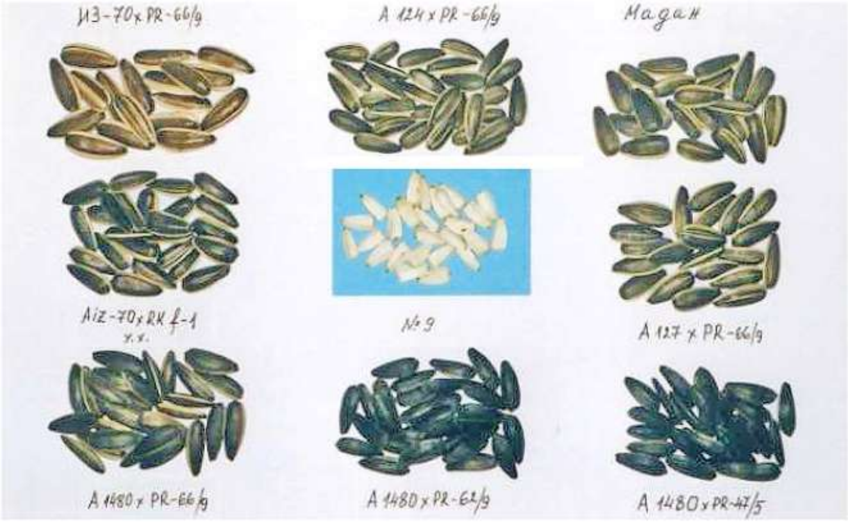

Figure 11: Seeds with a different colour.

Figure 12: Production of new heterosis hybrids.

mitochondria) is responsible for plant sterility. After confirming that there are no R*f* genes in the new plant material we can obtain sterile analogues (***A* lines**) of different *B* lines. After the BC_3 or BC_4 generations the study of the *general combining ability* of created *A*-lines can be initiated and after that, of their *specific combining ability* for developing the best new hybrid combinations. After a new hybrid (*A* line × *R* line) is created, it is tested and possibly introduced to the EAVTFISC in Bulgaria and similar authorized agencies of other countries.

***Cytoplasmic male sterility* (*CMS*)** is an inherited biological phenomenon, which is expressed in male sterility while preserving the female fertility of flowers and plants (Dankov 1973). The first stable source of CMS and practically the only one currently used in the heterosis selection of sunflowers is labelled PET1-CLASSICAL

Figure 13: B lines from the cross: a – *H. neglectus* × *H. annuus* (M-8-A1 B), b – *H. annuus* × *H. nuttallii* ssp. *rydbergii* (1073 B), c1 and c2 – (*H. hirsutus* × *H. annuus*) × *H. annuus*, and etc.

Figure 14: R lines from the cross: a – *H. annuus* × *H. bolanderi*, b – *H. annuus* × *H. giganteus* (2521 R), c – *H. annuus* × *H. neglectus* (3209 R), d – *H. annuus* × *H. nuttallii* ssp. *rydbergii* (line1 – 1018 R and line2 – 1070 R), e – *H. annuus* × *H. smithii*, f – *H. annuus* × *H. pauciflorus* ssp. *subrhomboideus* (line1 – 2879 R; line2 – 3261 R, and line3 – 42 R), g – *H. annuus* × *H. hirsutus* (line1 – Sc 98 R and line2 – 2548 R), h *H. eggertii* × *H. annuus*.

Table 2: List of Bulgarian CMS sources in sunflower (Serieys and Christov 2005).

Common designation	Origin species	Accession code	Obtain in generation	Author report	FAO code
AN-67	*H. annuus*	E-067	F_1	Christov (1992a)	ANN10
AN-58	*H. annuus*	E-058	F_6	Christov (1994a)	ANN11
AN-2-91	*H. annuus*	E-002	F_5	Christov (1991)	ANN12
AN-2-92	*H. annuus*	E-002	F_6	Christov (1992b)	ANN13
CMS-G	*H. annuus*	v. Gigant		Christov (1993)	ANN15
CMS-DP	*H. annuus*	p. DP-1 08		Christov (1993)	ANN16
CMS-VL	*H. annuus*	p.1638/4		Christov (1993)	ANN17
	H. annuus	GT-E-126		Nikolova and Christov (2004b)	ANN18
Hemus	*H. annuus*	Cms H		Christov (1993)	MUT1
Peredovick	*H. annuus*	Cms P-UZ		Christov (1993)	MUT2
Stadion	*H. annuus*	Cms S		Christov (1993)	MUT3
Peredovick	*H. annuus*	Cms P-114		Christov (1993)	MUT4
Peredovick	*H. annuus*	Cms P-92		Christov (1996a)	MUT5
Voronejskii	*H. annuus*	Cms VO 481		Christov (2002)	MUT6
Peredovick	*H. annuus*	Cms P 450 Gy		Christov (1994b)	MUT13
ARGOPHYLLUS	*H. argophyllus*	E-006	F_1	Christov (1990)	ARG1
ARGOPHYLLUS	*H. argophyllus*	E-007	F_1	Christov (1990)	ARG2
ARGOPHYLLUS	*H. argophyllus*	E-006	BC_1	Christov (1999b)	ARG3
ARG3-M1	*H. argophyllus*	E-006	new BC_1	Christov (2000)	ARG3-M1
ARGOPHYLLUS	*H. argophyllus*	E-007	BC_1F_7	Christov (1998a)	ARG4
DV-10	*H. debilis*	E-010	F_2	Christov (1994b)	DEB1
HIRSUTUS	*H. hirsutus*	HIR-29	BC_1	Hristova-Cherbadji (2004)	HIR1
PET-34	*H. petiolaris*	E-034	BC_1F_6	Christov (1991)	PET4
PHIR-27	*H. praecox* ssp. *hirtus*	E-027	F_2	Christov (1990)	PRH1
PPR-28	*H. praecox* ssp. *praecox*	GT-E-028		Nikolova (1999)	PRP2
RUN-29	*H. praecox* ssp. *runyoni*	E-029	F_4	(Christov 1999b)	PRR1
RIG-M-28	*H. rigidus*	M-028	BC_1F_2	Christov (1991)	RIG2
STRUMOSUS	*H. strumosus*	STRUM-56	BC_1F_5	Christov (1997)	STR1

CMS or “French cytoplasm”, and was discovered by Leclercq (1969). It originates from *H. petiolaris* Nutt. by using various methods 72 new CMS souses, out of which 28 are Bulgarian (Christov 1990, 1992a, b, 1993, 1999a, b; Serieys and Christov 2005) (Table 2), were discovered with the biggest amount being obtained thanks to remote hybridization. CMS attachment in the sunflower is controlled by a system with changing interactions between nuclear and cytoplasmic genes.

The carried out remote hybridization allows diverse hybrids and valuable starting material for the selection of sunflowers. Currently, some of these lines are certified and can be used practically in seed production for the creation of new varieties.

The diseases, pests and abiotic stress in plants are barriers to obtaining high and stable yields. The optimal conditions that are created for the cultivated sunflower are also very favourable for the development of new, more virulent races of pathogens and parasites, capable of attacking even the most resistant plants, due to the parallel course of a natural formative process in nature. For this reason, it is necessary to continue work on the most interesting new lines to maintain their sustainability.

The genetic potential of wild species from the genus Helianthus and other genera of the family Compositae is a natural and appropriate way to increase the variability of the cultivated sunflower. In the future, research can be continued with the transfer of genetic material from the still unused wild species, i.e. the realization of new crosses between different species.

The good results allow for future study by molecular methods of the genes that determine the traits of interest. The ultimate goal of such a study is their sequencing and easy tracking during the transfer from one material to another through *Marker Assistant Selection.*

In the last few years, the areas sown with convectional sunflower, which is less resistant to diseases, have been increasing. Another challenge is the drought that has been prevalent in recent years, which strongly affects the obtained yield. Both of these factors create a big opportunity for improvements that might be achieved by distant hybridization.

4 Conclusions

In Bulgaria, after a long period, various materials having a high combinative ability, complex resistance to diseases, the parasite Broomrape and some herbicides, as well as high amounts of oil have been obtained as a result of the use of efficient application of different selection methods. New sources of CMS and restoration of fertility genes (*Rf* genes) have also been discovered. Moreover, species that have never been crossed before have been successfully used to make new hybrids. There are still many forms that could be developed or studied, as well as never-ending opportunities for practical and commercial applications of the mentioned Bulgarian findings.

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