HELIA, **25**, Nr. 36, p.p. 65-78, (2002) UDC 633.854.78:631.527

USING THE COLLECTION OF WILD SPECIES IN SUNFLOWER BREEDING

N. Tavoljanski[1*], A. Yesaev[1], V. Yakutkin[2],
E. Akhtulova[1] and V. Tikhomirov[1]

[1]*"VIS, Ltd", Centralnaya 43 "a", Veidelevka, Belgorod region, 309720, Russia*
[2]*All-Russian Plant Protection Institute (VIZR), Shosse Podbelskogo 3, St-Petersburg, Pushkin 196620, Russia*

Received: November 08, 2001
Accepted: June 03, 2002

SUMMARY

We started to collect wild sunflower (*Helianthus* L.) species in 1998. The main task of this collection is to promote the development of new breeding material. In particular, it was planned to use the wild species as resistance sources to the major diseases of sunflower and as *Rf* genes sources to the new CMS types. The collection consists presently of 140 accessions of annual wild species and 110 samples of 27 perennial ones. The assessment of the wild sunflowers collection for resistance to *Sclerotinia* (*Sclerotinia sclerotiorum*), *Phomopsis* (*Phomopsis helianthi*) and broomrape (*Orobanche cumana*) was carried out on artificial infectious backgrounds. Resistance to other diseases was estimated simultaneously under natural infection. The immunologic potential of the collection was assessed three times during the vegetative period: at early stages of ontogeny (first week of June), at full flowering (second week of August) and at the end of the vegetation period (fourth week of September). A group of 101 samples showed various types of resistance to a complex of diseases. F_1 and F_2 generations were produced from more than 100 cross combinations of sunflower inbred lines and wild annual species. The tolerance to *Phomopsis* in field conditions was recorded in the F_1 and F_2 interspecific hybrids. Also, several sources of fertility restoration for new CMS types (ANN1, ARG1, PEF1, GIG1 and RIG1) have been found among the accessions of wild annual sunflower species. Thus, certain samples from the collection may be useful for breeding to disease resistance. The development of new initial material for sunflower breeding has already started.

Key words: **sunflower (*Helianthus* L.) wild species, interspecific hybrids, *Phomopsis* (*Phomopsis helianthi*), *Sclerotinia* (*Slerotinia sclerotiorum*) tolerance, fertility restoration of new CMS sources**

* Corresponding author, Phone: (07237) 5-50-83,
Fax: (07237) 5-50-66, e-mail: msch@mail.ru

INTRODUCTION

The Veidelevka Institute of Sunflower is the first private Russian enterprise to carry out plant breeding, seed production and seed sale of several crops, but primarily of sunflower. The breeding of varieties and hybrids of oilseed and confectionery sunflower proceeds in the following major directions:

- high level of GCA;
- productivity of female parent lines of hybrids;
- resistance or tolerance to downy mildew (*Plasmopara halstdii*), *Phomopsis* stem canker (*Phomopsis helianthi*), different forms of *Sclerotinia* wilt (*Sclerotinia sclerotiorum*), *Verticillium* wilt (*Verticillium dahliae*) and broomrape (*Orobanche cumana*);
- precocity.

Five oilseed hybrids and two varieties from our breeding programs have been registered and are now in wide cultivation in the central black-soil region of Russia.

Fungal diseases such as *Phomopsis* and *Sclerotinia* have presented an increasing danger to the sunflower production in our region in recent years. A continuous increase of sunflower cultivation area and the resulting rotation reduction will, apparently, further complicate the phytopathological situation in the region. The development of cultivars with genetic resistance is one of the essential factors of disease control. Wild sunflower species are of great interest as potential sources for the enrichment of the germplasm of cultivated sunflower.

Sources of genetic resistance to *Phomopsis* and downy mildew have been detected in both wild sunflower species and in lines developed from interspecific hybrids. Wild annual species such as *H. argophyllus*, *H. praecox*, *H. neglectus*, *H. petiolaris* and *H. debilis* are most frequently mentioned in scientific papers as resistant to *Phomopsis* and downy mildew (Beshard, 1997; Langar, 1998; Christov, 1999; Hahn, 1999; Seiler, 1999; Škorić, 1999; Romano, 1999). Wild species such as *H. argophyllus*, *H. praecox* and *H. paradoxus* have demonstrated a limited damage by the root and stem forms of *Sclerotinia* (Hahn, 1999a, 1999b; Seiler, 1999; Romano, 1999). The species *H. tuberosus* is considered as most prospective among the perennial wild sunflower species. Its resistance to *Phomopsis* and *Sclerotinia* has been reported (Langar, 1998; Hahn, 1999,1999b; Seiler, 1999); and the crossing ability of this species with annual sunflower is well known.

Of the sixty-two new CMS sources identified in sunflower so far, genes of fertility restoration (*Rf*) have been discovered for twenty-nine of them. Genetic analyses of the fertility restoration character have been conducted for sixteen CMS sources by various authors (overview by Serieys, 1999). These analyses demonstrated the dominant control of this character by one or two complementary genes.

The wild sunflower species collection was established in Veidelevka Institute of Sunflower in 1998. It is planned to use it for the development of sources of resistance to major diseases and for obtaining *Rf* sources for new CMS forms.

MATERIALS AND METHODS

Seed sets and pieces of rhizomes for the establishment of our collection were received from the following research centers: RPIS (Ames, USA), IFVC (Novi Sad, Yugoslavia), IWS "Dobroudja" (Bulgaria), research stations of VIR on Kuban and in Maykop (Russia), All-Russian Institute of Oil Crops (Krasnodar) and Institute of Oil Crops (Zaporozhie, Ukraine).

The seeds of wild sunflower species were germinated at the end of March – beginning of April. The seeds were placed in damp paper rolls at 0°C for one day and then at 25°C for 5-7 days; the seeds were scarified to facilitate germination. The germinated seeds were transferred into 200 ml pots filled with soil-sand mixture (2:1). The pots with the germinated seeds were kept in chambers for accelerated plant cultivation (illumination for twelve hours per day and temperature at 25°C). The obtained plantlets in the phase of 2-3 pairs of true leaves were sown into the infectious background (quarantine nursery) using the following planting schemes: 0.7 x 1.5 m for annuals and 4 x 4 m for perennial species, without replication.

Adding sclerotia into the soil (at the rate 5 sclerotia per 1 m^2) created the infectious background of *Sclerotinia*; the placement of infected vegetative debris at intervals between plants created the infectious background of *Phomopsis*. Additional infection of the samples by *Sclerotinia*, *Phomopsis* and other diseases by air-borne inoculum took place in due time.

Broomrape infestation was carried out before sowing by adding populations of broomrape seeds to the soil (1 g of seeds per 1 m^2). Broomrape seeds have been collected in sunflower fields of the Saratov and Belgorod regions. To estimate the obtained inoculum levels of the diseases and broomrape in the infectious backgrounds, a set of susceptible samples was used - the line HA 852 (very susceptible to *Sclerotinia*), the line HA 89 (susceptible to *Phomopsis*) and the variety the "Giant 549" (susceptible to broomrape). The line VIR 130 was used as the comparative standard for resistance to *Phomopsis*, the line VB 471 as the standard for resistance to broomrape in the given infectious background. Check plots were sown in two replications, thirty plants per replication.

The intensity of occurrence of the diseases and broomrape was expressed in percentage. The immunologic potential of the collection was checked three times during the vegetative period: at early stages of ontogeny (first week of June), at full flowering (second week of August) and at the end of the vegetation period (fourth week of September).

Interspecific hybridization of cultivated sunflower forms with wild annual and perennial sunflower species started in 1998. Reciprocal crosses between wild and cultivated forms of sunflower were made. Such forms of cultivated sunflower as the lines VB 4703, VB 471, VB 246 (parent lines of commercial hybrids), VIR 130 (GCA; *T, o, vs, p, M* genes), HA 335 (Pl_6 gene), HA 89, and also the variety "Fuxinka 62" (*T* gene) were included in the crossing scheme. CMS-analogues of the cultivated

lines were used in direct crosses, and the maintainer-analogues were used in reciprocal ones. It was expected that the foreign pollen would have advantage for fertilization due to strict self-incompatibility of the wild species. Besides, in the case of hybridization failure, the plants of parental wild species could be clearly distinguished from interspecific hybrids and removed.

Individual heads of wild and cultivated parents were isolated with bags 2-3 days before flowering. At flower, pollen mixture from sample was placed on the stigmata of the female heads. The pollination was repeated two-three times.

The estimation of the F_1 interspecific generation for the presence/absence of *Phomopsis* symptoms was carried out in conditions of natural infection. Sixty-four F_1 hybrid combinations were checked in the year 2000. Each combination included from 15 to 45 plants, without replications. Sixty F_1 hybrid combinations were checked in 2001; two replications with thirty plants in each, were obtained.

In 1998 we received six new CMS sources from All-Russian Institute of Oil Crops, Russia, and INRA, France: ANN-1, ARG-1, PEF-1, RIG-1, GIG-1 and MAX-1. We searched the wild species collection for sources of fertility restoration of these CMS types. The male-sterile inflorescences of the CMS sources were pollinated by pollen grains from wild species. The method of crossing was similar to the one mentioned above. Sixty-two cross combinations of were obtained.

RESULTS AND DISCUSSION

In the years 1999 and 2000, the collection of wild sunflower species of Veidelevka Institute of Sunflower numbered eight annual species (68 and 140 accessions, respectively) and twenty-seven perennial species (127 and 110, samples respectively) (Table 1).

In the year 2000, the immunology assessment of the annual sunflower species revealed the presence of seven diseases: *Phomopsis* stem canker (*Phomopsis helianthi*), *Sclerotinia* wilt (*Sclerotinia sclerotiorum*), *Verticillium* wilt (*Verticillium dahliae*), downy mildew (*Plasmopara halstedii*), rust (*Puccinia helianthi*), *Fusarium* (*Fusarium moniforme*) and *Phoma* black stem (*Phoma macdonaldii*). The presence of broomrape (*Orobanche cumana*) was recorded too.

One-hundred-and-seven accessions of wild annual sunflowers (76% of a collection) revealed disease symptoms. The frequency of disease incidence was as follows:

- *Phomopsis* was recorded on 76 samples (54% of a collection): 56 samples of *H. annuus*, 8 samples of *H. praecox*, 5 samples of *H. petiolaris*, 3 samples of *H. debilis* and 4 samples of *H. argophyllus*;
- the presence of broomrape was recorded on 56 samples (40% of a collection): 50 samples of *H. annuus*, 2 samples of *H. praecox* and 4 samples of *H. argophyllus*;
- *Verticillium* wilt was found on 17 samples (12 % of a collection): 6 samples of

Table 1: Accessions in the collection of wild sunflower species (*Helianthus* L.) at Veidelevka Institute of Sunflower – 1999, 2000

	No.	Species	Accession quantity	
			In 1999	In 2000
Annual	1	*H. annuus*	28	81
	2	*H. argophyllus*	9	9
	3	*H. bolanderi*	1	1
	4	*H. debilis*	4	9
	5	*H. neglectus*	4	5
	6	*H. niveus*	1	2
	7	*H. petiolaris*	12	24
	8	*H. praecox*	9	9
Perennial	1	*H. angustifolius*	1	1
	2	*H. decapetalus*	3	3
	3	*H. divaricatus*	5	9
	4	*H. eggertii*	1	1
	5	*H. floridanus*	1	1
	6	*H. giganteus*	5	5
	7	*H. grosseserratus*	10	10
	8	*H. hirsutus*	4	6
	9	*H. laetiflorus*	1	1
	10	*H. laevigatus*	1	1
	11	*H. macrophillus*	3	3
	12	*H. maximilliani*	5	6
	13	*H. mollis*	8	7
	14	*H. multiflorus*	2	2
	15	*H. nuttallii*	11	11
	16	*H. occidentalis*	4	2
	17	*H. pauciflorus*	1	1
	18	*H. resinosus*	1	1
	19	*H. rigidus*	4	5
	20	*H. salicifolius*	3	3
	21	*H. scaberimus*	1	1
	22	*H. simulans*	1	2
	23	*H. smithii*	1	1
	24	*H. strumosus*	5	3
	25	*H. subcanescens*	1	1
	26	*H.trachelifolius*	1	1
	27	*H. tuberosus*	22	22
		Interspecific hybrids of perennial types from Maykop Research Station	21	-

H. petiolaris, 9 samples of *H. annuus* and 2 samples of *H. argophyllus*;
- *Sclerotinia* stem and root rot were found on 16 samples (11% of a collection): 13 samples of *H. annuus*, 1 sample of *H. praecox* and 2 samples of *H. argophyllus*;
- rust was recorded on 15 samples (10% of a collection): 14 samples of *H. annuus* and 1 sample of *H. petiolaris*;
- *Fusarium* was recorded on 8 samples (5% of a collection): 6 samples of *H. annuus* and 2 samples of *H. praecox*;
- downy mildew was found on 3 samples (2% of a collection): 2 samples of *H. annuus* and 1 sample of *H. neglectus*;
- *Phoma* black stem was recorded on 1 sample of *H. annuus*.

The infection potential of *Phomopsis* and the infestation potential of broomrape were high with the samples from the wild sunflowers collection. The presence of *Phomopsis* on the susceptible line HA89 was 100%, while it did not exceeded 0.3% on the resistant line VIR 130. The incidence of broomrape on the susceptible variety Giant 549 reached 100%, with the average density of parasites of 20.3±1.33 per host plant. The occurrence of broomrape was not recorded on the control line VB 471. The infectious background of *Sclerotinia* was weak due to insufficient humidity. The damage of heads by the disease was not registered; the incidence of root and stem forms of *Sclerotinia* on the susceptible line HA 852 did not exceed 45%.

Of the samples of wild annual sunflowers tested in conditions of VIS, 24 had been investigated earlier in Yugoslavia (Škorić, 1999). A comparative analysis on resistance to *Phomopsis*, *Phoma* and broomrape showed the following (Table 2): identical levels of resistance/susceptibility to the above diseases and broomrape were found in 6 out of 24 samples. Partial concurrence of estimates was registered in 15 samples, and opposite estimates in 3 samples. There were no correlation between the results of our estimation and the results of Škorić (1999) for either one of the three pests. The correlation coefficient for all diseases was 0.18± 0.21 (Table 2).

We suppose that the established differences in resistance to the diseases and broomrape are due to ecological factors, genetic heterogeneity of the investigated populations of wild annual sunflowers, different infection potentials of the pathogens, and also due to probable differences in virulence among the pathogen populations. However, although the existence of broomrape races is well known, the question about the existence of *Phomopsis* races in Europe remains open (Langar, 1999; Maširević, 1999).

The ratio of resistant vs. susceptible samples in our research and in Yugoslavia was approximately identical. However, our estimates differed appreciably from the results obtained in Yugoslavia for *Phoma macdonaldii*. While 63% of the samples investigated in Yugoslavia were infected by *Phoma* black stem, we recorded *Phoma*

infection in only one sample of *H. annuus*. Potential sources of resistance to *Phoma* have not been identified yet in the annual wild species (Seiler, 1999).

Table 2: Comparison of results from Yugoslavia (Škorić, 1999) and Veidelevka, Russia – 2000, for resistance to *Phomopsis*, *Phoma* and broomrape in 24 samples of wild annual sunflower species

No.	Species	Accession No.	Resistance / susceptibility					
			Phomopsis helianthi		*Phoma macdonaldi*		*Orobanche cumana*	
			I*	II**	I	II	I	II
1	*H. annuus*	1963	MS***	R	S	R	R	R
2	*H. annuus*	1970	MS	S	R	R	R	S
3	*H. annuus*	2123	S	R	S	R	S	R
4	*H. annuus*	2128	MS	S	S	R	S	S
5	*H. annuus*	2136	MS	S	S	R	R	S
6	*H. annuus*	2150	MS	S	S	R	S	S
7	*H. annuus*	2162	MS	R	S	R	R	S
8	*H. annuus*	2165	MS	R	S	R	R	R
9	*H. annuus*	2170	MS	S	R	R	R	R
10	*H. annuus*	2171	MS	S	S	R	S	R
11	*H. annuus*	2173	MS	S	S	R	R	S
12	*H. annuus*	2187	MS	R	S	R	S	S
13	*H. annuus*	2188	MS	R	S	R	S	S
14	*H. petiolaris*	338	MS	S	R	R	R	R
15	*H. petiolaris*	722	R	R	R	R	R	R
16	*H. petiolaris*	1383	MS	S	R	R	R	R
17	*H. petiolaris*	1910	R	R	R	R	R	R
18	*H. petiolaris*	2009	MS	R	S	R	R	R
19	*H. petiolaris*	2113	MS	R	R	R	S	R
20	*H. petiolaris*	2119	MS	R	R	R	R	R
21	*H. petiolaris*	2126	MS	R	S	R	R	R
22	*H. petiolaris*	2178	MS	R	S	R	S	R
23	*H. petiolaris*	2203	MS	S	S	R	S	R
24	*H. praecox*	1801	R	S	R	R	R	R
Correlation coefficient for individual disease			R=0.09±0.21		-		r=0.18±0.21	
Correlation coefficient for all diseases			r=0.18±0.21					

Note: * Results from Novi Sad, as reported by Škoriæ, 1999; ** results of estimation in Veidelevka; *** R-resistance (noted “0”-“1” by Škoriæ, 1999); MS- medium susceptibility (noted “3” by Škoriæ, 1999); S - susceptibility (noted “4” by Škoriæ, 1999)

It is possible that the low level of *Phoma* presence on the samples of annual wild species from our collection is due to low pathogen concentration in the environmental conditions of our experiment. However, we recorded 100% presence of *Phoma* in some cultivated sunflower lines grown in a quarantine nursery in the year 2000. That is why we believe that the resistance of annual wild species to *Phoma* needs further research.

The immunologic screening of the wild annual sunflower collection under the regional conditions of Russia - at Veidelevka Institute of Sunflower in the Belgorod area - showed no external symptoms of the diseases and damage of root system by broomrape in 35 of the investigated samples (Table 3).

Table 3: Wild annual sunflower species which were free from any disease symptom, Veidelevka Institute of Sunflower, 2000

No.	Species	Accession
1	*H. annuus*	ANN-1064
2	*H. annuus*	ANN-1173
3	*H. annuus*	ANN-1672
4	*H. annuus*	ANN-2093
5	*H. annuus*	ANN-2123
6	*H. annuus*	ANN-2165
7	*H. annuus*	ANN-2294
8	*H. bolanderi*	E-009
9	*H. debilis*	DEB-014
10	*H. debilis*	DEB-1218
11	*H. debilis*	DEB-1569
12	*H. debilis*	DEB-1564
13	*H. debilis*	DEB-1675
14	*H. debilis*	?
15	*H. neglectus*	NEG-465
16	*H. neglectus*	NEG-461
17	*H. neglectus*	NEG-460
18	*H. neglectus*	?
19	*H. niveus*	NIV-639
20	*H. niveus*	NIV-641
21	*H. petiolaris*	PET-13
22	*H. petiolaris*	PET-722
23	*H. petiolaris*	PET-1264
24	*H. petiolaris*	PET-1441
25	*H. petiolaris*	PET-1778
26	*H. argophyllus*	ARG-1834
27	*H. petiolaris*	PET-1910
28	*H. petiolaris*	PET-2009
29	*H. petiolaris*	PET-2105
30	*H. petiolaris*	PET-2113
31	*H. petiolaris*	PET-2119
32	*H. petiolaris*	PET-2178
33	*H. petiolaris*	PET-2306
34	*H. petiolaris*	PET-586919
35	*H. praecox*	PRA-560400

This group included 2 samples of *H. niveus* (100% of samples of this species), 1 sample of *H. bolanderi* (100%), 4 samples of *H. neglectus* (80%), 6 samples of *H. debilis* (67%), 13 samples of *H. petiolaris* (54%), 1 sample of *H. praecox* (11%), 1 sample of *H. argophyllus* (11%) and 7 samples of *H. annuus* (9%). These results are in agreement with the results of Škorić (1999).

In the course of two-year screening for resistance to natural *Phomopsis* infection, which included 120 F_1 and F_2 interspecific combinations between cultivated sunflower and wild annual species, 46 cross combinations showed no symptoms of the disease. Those were the following species:

- *H. annuus*, including samples H-29, H-36, H-41, H-94, H-151, H-181, ANN-155, ANN-376, ANN-529, ANN-1173, ANN-1389 and ANN-2168;
- *H. argophyllus*, including samples ARG-1805, ARG-1806, ARG-1807, ARG-1812, ARG-1820, ARG-545665 and ARG-Mozambik;
- *H. petiolaris*, including samples PET-815, PET-1441, PET-1910, PET-2011, PET-2113 and PET-2203;
- *H. neglectus*, including samples NEG-1182, NEG-460 and NEG-461;
- *H. praecox*, including samples PRA-416 and PRA-560400.

Sixteen samples demonstrated tolerance to *Phomopsis* during the test on an infectious background in 2000; nine samples were susceptible in this test; the other five samples were omitted from the test (Table 4). F_1 crosses between cultivated sunflower and some wild species susceptible to *Phomopsis* have shown resistance to the disease, while some crosses with resistant wild species (10%) have appeared susceptible to *Phomopsis*. We believe that this phenomenon is connected with the polygenic control of resistance or susceptibility to *Phomopsis* in sunflower. Similar results were reported by Beshard (1997). Our investigation showed what there is an essential connection in the transfer of resistance to *Phomopsis* from wild annual species to interspecific crosses; in our experiment, the value of this connection reached $r=0.45\pm0.20$.

When placed on the infectious background, most samples of perennial sunflowers from our collection did not show symptoms of the major diseases. *Erysiphe cichoracearum* was registered on 41 samples and *Verticillium* wilt on 7 samples. Sixty-six samples from 22 species showed no disease symptoms: 3 samples of *H. decapetalus* (100% of the samples of this species), 6 samples of *H. divaricatus* (67%), 2 samples of *H. giganteus* (40%), 7 samples of *H. grosseserratus* (70%), 3 samples of *H. hirsutus* (50%), 1 sample of *H. laevigatus*, 2 samples of *H. macrophillus* (66%), 4 samples of *H. maximilliani* (66%), 3 samples of *H. mollis* (42%), 1 sample of *H. multiflorus* (50%), 5 samples of *H. nuttallii* (45%), 2 samples of *H. occidentalis* (100%), 1 sample of *H. pauciflorus*, 1 sample of *H. resinosus*, 4 samples of *H. rigidus* (80%), 3 samples of *H. salicifolius* (100%), 1 sample of *H. scaberimus*, 2 samples of *H. simulans* (100%), 2 samples of *H. strumosus* (66%), 1 sample of *H. subcanescens*, 1 sample of *H. trachelifolius*, 11 samples of *H. tuberosus* (50%).

Table 4: Cross combinations which were free of *Phomopsis* symptoms under conditions of natural infectious background, Veidelevka Institute of Sunflower, 2000, 2001

No.	Generation	Year of screening	Parents		
			Female	Male	
			Species/line	Species/line	Accessions
1	F_1	2000	VB 471	*H. annuus*	*H-29**, H-94, *ANN-376*, ANN-1173**
2	F_1	2000	-"-	*H. argophyllus*	ARG-1807
3	-"-	-"-	-"-	*H. neglectus*	NEG-1182
4	-"-	-"-	-"-	*H. petiolaris*	PET-2203
5	-"-	-"-	VB 4703	*H. annuus*	H-36, *ANN-529*
6	-"-	-"-	-"-	*H. argophyllus*	ARG-1812
7	-"-	-"-	-"-	*H. neglectus*	NEG-460, NEG-461
8	-"-	-"-	HA 89	*H. annuus*	*H-29*, H-36, *H-41*, H-181, ANN-155, *ANN-376*, *ANN-529*
9	-"-	-"-	-"-	*H. argophyllus*	ARG-1806
10	-"-	-"-	-"-	*H. petiolaris*	PET-815, *PET-2011*, PET-2113,
11	-"-	-"-	-"-	*H. praecox*	PRA-416
12	F_2	2000	VB 471	*H.argophyllus*	ARG-1806
13	F_2	2000	*H. argophyllus* ARG-1806	Fuxinka 62	--
14	-"-	-"-	ARG-1805	Fuxinka 62	--
15	-"-	-"-	*H. annuus* ANN-1173	Fuxinka 62	--
16	-"-	-"-	ANN-1389	Fuxinka 62	--
17	-"-	-"-	Í-94	Fuxinka 62	--
18	F_1	2001	VIR 130	*H. annuus*	*ANN-376*, ANN-2168,
19	F_1	2001	-"-	*H. argophyllus*	ARG-1805, ARG-1806, ARG1807, ARG-1820
20	-"-	-"-	VB 246	*H. annuus*	H-151
21	-"-	-"-	-"-	*H. argophyllus*	ARG-1805, ARG-1806, ARG-1807, ARG-545665, Mozamb.,
22	-"-	-"-	-"-	*H. petiolaris*	PET-1441, PET-1910, PET-2203
23	-"-	-"-	-"-	*H. praecox*	PRA-560400

* The accessions marked as *H-29* were susceptible to *Phomopsis* on artificial background of the 2000 disease trial.

** The accessions underlined as ANN-1173 were tolerant to *Phomopsis* in artificial background of the 2000 disease trial.

A greater part of samples of the collection of perennial sunflowers showed multiple resistance to the major pathogens of sunflower. However, our attempts to cross perennial species with cultivated sunflower have not been successful. The majority of perennial samples begin to flower in late autumn; only some fifteen to twenty per-

ennial samples bloom in summer. Thus, we cannot make as many crosses with perennial species as would be desirable. Furhermore, the few seeds that form at the end of summer cannot achieve physiological maturity. This problem will be solved by resorting to the method of *in vitro* culture of immature embryos.

Sixty-two crosses were made in 1999 between wild perennial sunflowers and six new CMS sources. The analysis of the F_1 hybrids revealed the presence of fertile plants in 21 cross combinations. The following samples restored fertility of the new CMS sources: CMS ANN1 was restored by H-98, H-94, ARG-1807, ARG-1812, NEG-1182; CMS ARG1 was restored by H-29, PET-2203; CMS PEF1 was restored by ANN-529, H-36, ARG-1812, NEG-1182; CMS GIG1 was restored by ANN-529, H-28, ARG-1806; CMS RIG was restored by ANN-529, H-216, PET-815, PRA-1295, PRA-1828, NEG-1182 (Table 5).

Table 5: Results of interspecific crossing between new cms sources and wild annual sunflower species, F_1 generation, Veidelevka Institute of Sunflower, 2000

Female parent	Male parent		Male fertility/sterility
	Species	Accession	
CMS ANN-1	*H. annuus*	H-98, H-94	F/S *
--"--	*H. argophyllus*	Arg1807, Arg1812	F/S, F
--"--	*H. neglectus*	NEG-1182	F/S
CMS ARG-1	*H. annuus*	H-29	F
--"--	*H. petiolaris*	PET-2203	F/S
CMS PEF-1	*H. annuus*	ANN-529, H-36	F, F/S
--"--	*H. argophyllus*	ARG-1812	F/S
--"--	*H. neglectus*	NEG-1182	F/S
CMS GIG-1	*H. annuus*	ANN-529, H-28	F/S
--"--	*H. argophyllus*	ARG-1806	F/S
CMS RIG-1	*H. annuus*	ANN-529, H-216	F/S, F
--"--	*H. petiolaris*	PET-815	F
--"--	*H. praecox*	PRA-1295, PRA-1828	F/S, F
--"--	*H. neglectus*	NEG-1182	F/S

* Note: F/S –Fertile and sterile plants present in the F_1 population.

The sources of fertility restoration that we obtained have already been reported (the review of works, Serieys, 1999); the genetic control of fertility restoration of CMS RIG1 and CMS PEF1 has been described. Thus, our results supplement the list of known sources of fertility restoration in CMS ANN1, ARG1, REF1, GIG1 and RIG1.

CONCLUSIONS

Multiple resistance to diseases was found in the following populations of annual wild species: *H. niveus*, *H. neglectus*, *H. debilis*, *H. petiolaris*, *H. argophyllus*. Due to good crossing ability with cultural forms, wild annual species of sunflower repre-

sent an easily available reserve of genetic diversity for sunflower breeding. The development of new forms for sunflower breeding for resistance to *Phomopsis* has started. The polygenic control of this character forces us to resort to convergent crossing in order to attain the final goal – hybrids with tolerance to *Phomopsis*.

Annual species such as *H. annuus* (7 samples), *H. petiolaris* (2 samples), *H. argophyllus* (3 samples), *H. praecox* (2 samples) and *H. neglectus* (1 sample) were found to possess genes for fertility restoration in the new CMS sources ANN1, ARG1, PEF1, GIG1 and RIG1.

ACKNOWLEDGEMENTS

We would like to thank all research centers which have kindly provided us with sets of seeds and rhizomes for the establishment of our collection of wild sunflower species: research stations of VIR on Kuban and in Maykop (Russia), All-Russian Institute of Oil Crops (Krasnodar, Russia), Institute of Oil Crops (Zaporozhie, Ukraine), IWS "Dobroudja" (Bulgaria), IFVC (Novi Sad, Yugoslavia) and RPIS (Iowa, Ames, USA).

UTILIZACION DE LA COLECCION DE ESPECIES SILVESTRES EN LA SELECCION DEL GIRASOL

RESUMEN

Por la coleccion de especies silvestres del girasol (*Helianthus* L.) hemos empezado en 1998. La tarea basica de esta coleccion es la promocion del trabajo para la creacion de nuevo material selectivo. Fue particularmente planeado que las especies silvestres se utilicen como fuentes de resistencia a las enfermedades de girasol predominantes y como fuentes del gen *Rf* para nuevos tipos de la esterilidad masculina citoplasmatica. La coleccion consiste por el momento en 140 muestras de las especies silvestres de un ano y en 110 muestras de 27 especies silvestres de varios anos. La evaluacion de la coleccion de especies silvestres con respecto a la resistencia a la esclerocina (*Sclerotinia sclerotiorum*), fomopsis (*Phomopsis helianthi*) y orobanca (*Orobanche cumana*) fue hecha en el fondo infectado artificial. Al mismo tiempo fue hecha la investigacion de la resistencia a otras enfermedades en las condiciones de la infeccion natural (aerogenica). El potencial inmunologico de la coleccion era evaluado tres veces durante el periodo de vegetacion: en las fases tempranas de la ontogenesis (primera semana del mes de junio), en el florecimiento completo (segunda semana del mes de agosto), y al fin del periodo de vegetacion (cuarta semana del mes de septiembre). Un grupo de 101 muestras indico diversos tipos de la resistencia al complejo de enfermedades. Han sido producidas las generaciones F_1 y F_2 de mas de 100 combinaciones de cruce de las lineas consaguineas del girasol cultivado y de las especies silvestres de un ano. Fue constatada la presencia de la tolerancia de *Phomopsis* en la interspecies F_1 y F_2 de los hibridos cultivados en las condiciones de campo. Entre las especies silvestres de un ano fueron encontradas algunas fuentes de restauracion de la fertilidad para los nuevos tipos de CMS (ANN1, ARG1, PEF1, GIG1 y RIG1). Eso signifa que ciertas muestras de la coleccion pueden ser utiles en la selec-

cion con respecto a la resistencia a las enfermedades. Empezo ya la creacion del nuevo material inicial para la seleccion del girasol.

UTILISATION DE LA COLLECTION D'ESPÈCES SAUVAGES DANS LA CULTURE DU TOURNESOL

RÉSUMÉ

Nous avons commencé à collectionner des espèces sauvages de tournesol (*Helianthus* L.) en 1998. Le but principal de ce travail est de faire avancer la création de nouveau matériel de culture. L'objectif est surtout l'utilisation d'espèces sauvages comme sources de résistance envers les maladies les plus importantes du tournesol et comme sources de gène *Rf* pour les nouveaux types de stérilité cytoplasmique mâle (CMS). La collection est actuellement composée de 140 échantillons d'espèces sauvages annuelles et de 110 échantillons de 27 espèces sauvages vivaces. La résistance de la collection d'espèces sauvages à la *Sclerotinia* (*Sclerotinia sclerotiorum)* au *Phomopsis* (*Phomopsis helianthi)* et à l'orobanche (*Orobanche cumana)* a été évaluée dans un environnement artificiellement infecté. La résistance à d'autres maladies a été évaluée en même temps dans des conditions d'infection naturelle (aérogénique). Le potentiel d'immunologie de la collection a été évalué trois fois au cours de la période de végétation: dans les phases précoces d'ontogenèse (première semaine du mois de juin), en pleine floraison (deuxième semaine du mois d'août), et à la fin de la période de végétation (quatrième semaine du mois de septembre). Un groupe de 101 échantillons a montré différents types de résistance à un ensemble de maladies. Des générations F_1 et F_2 ont été produites à partir de plus de 100 combinaisons de croisements de lignes inbred de tournesol et d'espèces sauvages annuelles. On a constaté la présence de tolérance au *Phomopsis* chez les hybrides interspecies F_1 et F_2 cultivés dans les champs. Parmi les espèces sauvages annuelles, on a aussi trouvé quelques sources de rétablissement de la fertilité pour les nouveaux types CMS (ANN1, ARG1, PEF1, GIG1 et RIG1). Cela signifie que certains échantillons de la collection peuvent être utiles pour la culture faite en vue d'établir une résistance envers les maladies. La création de nouveau matériel initial pour la culture du tournesol est déjà commencée.