

## RESISTANCE TO DISEASES, OBTAINED THROUGH INTERSPECIFIC HYBRIDIZATION

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### SUMMARY

Two accessions of the wild species *Helianthus annuus* L., GT-E-112 and GT-E-126, carried genes for resistance to *Plasmopara helianthi* Novot., *Phomopsis helianthi* Munt.-Cvet. *et al.* and *Orobanche cumana* Wallr. The material produced by interspecific hybridization with susceptible cultivated sunflower showed resistance to the three pathogens. Some progenies were resistant to two pathogens simultaneously. Self pollination helped to increase the percentage of resistance up to 100%. Accessions GT-E-112 and GT-E-126 of the wild species *Helianthus annuus* could be successfully used as donors for resistance to *Plasmopara helianthi*, *Phomopsis helianthi* and *Orobanche cumana*.

**Key words:** interspecific hybridization, wild *Helianthus annuus*, *Plasmopara helianthi*, *Orobanche cumana*, resistance

### INTRODUCTION

Diseases are limiting factor of production in the majority of sunflower-growing countries. The cultivated sunflower has a narrow genetic base and it is deficient in resistance genes. Sources of resistance have been found in wild sunflowers (Škorić, 1988). According to Thompson *et al.* (1978) and Christov (1990) some accessions of *Helianthus annuus* carry genes for resistance to *Plasmopara helianthi* Novot. Lines have been identified with tolerance to *Phomopsis helianthi* Munt.-Cvet. *et al.*, originating from *Helianthus annuus* accessions. The main sources for genetic resistance to *Orobanche cumana* Wallr. originated from *Helianthus tuberosus* (Seiler, 1992).

The aim of our study was to investigate two different accessions of the wild species *Helianthus annuus* as potential sources of resistance to two major diseases, *Plasmopara helianthi* and *Phomopsis helianthi*, and the parasite *Orobanche cumana*.

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## MATERIALS AND METHODS

The investigations were carried out at the Institute for Wheat and Sunflower "Dobroudja" during 1997-1998.

Two accessions of the annual diploid species *Helianthus annuus* - GT-E-112 and GT-E-126 - were included in the study together with the cultivated sunflower (*H. annuus*) inbred lines 1607, 2607, 1234, 3064, HA-300 and HA-821 and opv. Peredovik.

Twenty plants were studied in every accession and selection number. Self pollination was applied.

The evaluation of the material for downy mildew resistance (*Plasmopara helianthi* Novot.) was made according to the standard method (Vear and Tourvieille, 1987). The evaluation of the material for broomrape resistance (*Orobanche cumana* Wallr.) was carried out in greenhouse conditions according to the standard method (Panchenko, 1975). The inoculation for *Phomopsis* evaluation (*Phomopsis helianthi* Munt.-Cvet. *et al.*) was made according to the method of Tourvieille *et al.* (1988) in an infection plot with additional irrigation.

Oil content was determined by the method of nuclear magnetic resonance.

## RESULTS AND DISCUSSION

The preliminary evaluation of the annual wild species of *Helianthus annuus*, accessions GT-E-112 and GT-E-126, showed that both accessions carried genes for resistance to *Plasmopara helianthi* (25-30%), *Phomopsis helianthi* (25%) and *Orobanche cumana* (20-33%). The sunflower inbred lines and the variety Peredovik included in the investigation were susceptible. As a result of the interspecific hybridization between the wild *H. annuus* and the cultivated *H. annuus* hybrid material was produced and evaluated in the early F<sub>2</sub>, F<sub>3</sub>, F<sub>4</sub> and F<sub>5</sub> generations (Table 1).

### **Resistance to *Plasmopara helianthi* Novot.**

The obtained interspecific hybrids were studied for their reaction to *Plasmopara helianthi* (Table 1a). The value of resistance varied from 15 to 100%. The variation in the degree of resistance to mildew was due to the fact that the wild species were maintained as populations and the hybridization was carried out with pollen mixture. Thus, as a result of segregation, various degrees of resistance to *Plasmopara helianthi* were obtained in F<sub>2</sub>, F<sub>3</sub> and F<sub>4</sub> generations. Christov (1990, 1996), Christov, Shindrova and Entcheva (1996) claimed that resistance could be increased up to 100% in subsequent generations by self pollination. That is why we selfed plants showing a certain percent of resistance to mildew and studied their reaction to the pathogen in the following year. It could be seen from the results in

Table 1b that even one year of selfing could help to increase the resistance to *Plasmopara helianthi* up to 100% in the next generations.

Table 1: Evaluation of reaction of interspecific hybrids in early generations to *Plasmopara helianthi*

Origin	Selection No.	Generation	Resistance to <i>Pl. helianthi</i> (%)	Seed oil content (%)	Days to maturity
1607A x E-126	778/97	F <sub>2</sub>	50.0	49.69	101
1607A x E-112	808/97	F <sub>2</sub>	75.0	35.14	98
2607A x E-112	809/97	F <sub>2</sub>	55.6	45.30	111
1607B x E-126	1051/97	F <sub>3</sub>	17.5	43.00	101
2607A x E-126	1175/97	F <sub>3</sub>	62.5	52.04	101
3064A x E-126	1042/97	F <sub>3</sub>	58.6	51.16	115
HA-821B x E-126	1185/97	F <sub>3</sub>	71.4	47.20	126
1234A x E-112	1058/97	F <sub>3</sub>	15.0	39.98	101
HA-300A x E-112	1063/97	F <sub>3</sub>	94.5	42.13	117
HA-300B x E-112	1064/97	F <sub>3</sub>	70.0	50.09	117
E-112 x HA-821	1068/97	F <sub>3</sub>	23.6	49.82	117
1607B x E-126	1697/97	F <sub>4</sub>	50.0	42.83	114
1234A x E-126	1040/97	F <sub>4</sub>	100.0	46.29	97
3064A x E-112	1612/97	F <sub>4</sub>	56.4	41.89	98

Table 1b: Reaction of resistant hybrid material to *Plasmopara helianthi* as a result of self-pollination

Origin	Selection No.	Generation	Resistance %	Selection No.	Generation
1607A x E-126	778/97	F <sub>2</sub>	50.0	870/98	F <sub>3</sub>
2607A x E-112	809/97	F <sub>2</sub>	55.6	903/98	F <sub>3</sub>
2607A x E-126	1175/97	F <sub>3</sub>	62.5	1048/98	F <sub>4</sub>
3064A x E-126	1042/97	F <sub>3</sub>	58.6	1065/98	F <sub>4</sub>
HA-300A x E-112	1063/97	F <sub>3</sub>	94.5	1122/98	F <sub>4</sub>
HA-300B x E-112	1064/97	F <sub>3</sub>	70.0	1123/98	F <sub>4</sub>

#### Resistance to *Orobanche cumana* Wallr.

The reaction of the hybrid material to *Orobanche cumana* is presented in Table 2. The percentage of resistance varied from 10 to 100%. This variation was due to the same fact as pointed out above, i.e., to the segregation in the early generations. According to Tsvetkova and Shindrova (1987) the percentage of resistance to *Orobanche cumana* could reach 100% in the following generations as a result of selfing. Unfortunately our results from 1998 showed that in two progenies of 1.1234 A x E-112 and 1.2607 A x E-126 the resistance of 10% and 11.1%, respectively, was lost in the evaluated plants. Nevertheless our results with hybrid material produced from perennial species (Nikolova, Christov and Shindrova, 1998) confirmed the assumption of Tsvetkova and Shindrova. That is why the investigations on this matter should continue.

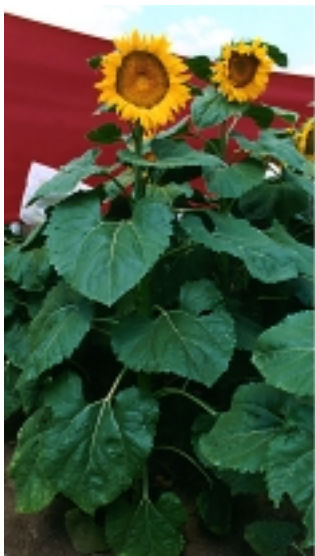


Figure 1:  $F_4$  1.1607 A x *H. annuus* (E-126)  
(100% resistant to *Plasmopara helianthi*)



Figure 2:  $F_5$  1.3064 A x *H. annuus* (E-126)  
(100% resistant to *Plasmopara helianthi*)



Figure 3:  $F_6$  opv. Peredovik x *H. annuus* (E-112)  
(75% resistant to *Phomopsis helianthi*)



Figure 4:  $F_5$  1.HA-300 A x *H. annuus* (E-112)  
(100% resistant to *Plasmopara helianthi*;  
70% resistant to *Orobanche cumana*)



Figure 5:  $F_4$  opv. Peredovik x *H. annuus* (E-126)  
(50% resistant to *Phomopsis helianthi*;  
25% resistant to *Orobanche cumana*)

Table 2: Evaluation of the reaction of interspecific hybrids in early generations to *Orobanche cumana*

Origin	Selection No.	Generation	Resistance to <i>O. cumana</i> (%)	Seed oil content (%)	Days to maturity
Peredovik x E-126	668/97	F <sub>2</sub>	25.0	50.94	108
1234A x E-112	1058/97	F <sub>3</sub>	10.0	39.98	101
HA-300A x E-112	1063/97	F <sub>3</sub>	70.0	42.13	117
E-112 x HA-821	1068/97	F <sub>3</sub>	100.0	49.82	117
2607A x E-126	1175/97	F <sub>3</sub>	11.1	50.04	101
1234A x E-112	1469/97	F <sub>4</sub>	37.5	40.00	99
E-126 x 2607	1592/97	F <sub>4</sub>	87.5	32.90	98
E-126 x 2607	1710/97	F <sub>4</sub>	22.2	40.75	114

**Resistance to *Plasmopara helianthi* Munt.-Cvet. et al.**

The hybrid material was also studied for its reaction to *Phomopsis helianthi*. For this evaluation hybrid progenies were selected that showed no infection in field conditions. Thus they were tested for resistance to *Phomopsis* in an infection plot using a method of inoculation. The results are given in Table 3. They confirmed the presence of resistance in most of the material. As a result of this study we could also claim that the annual species *H. annuus* carried genes for resistance to *Phomopsis helianthi*.

Table 3: Evaluation of reaction of interspecific hybrids in early generations to *Phomopsis helianthi*

Origin	Selection No.	Generation	Resistance to <i>P. helianthi</i> (%)	Seed oil content (%)	Days to maturity
HA-300A x E-126	660-97	F <sub>2</sub>	100	-	109
Peredovik x E-126	668/97	F <sub>2</sub>	50	-	107
HA-300B x E-112	927/97	F <sub>3</sub>	75	44.50	113
E-112 x HA-821	930/97	F <sub>3</sub>	50	35.98	100
1607B x E-126	1434/97	F <sub>4</sub>	75	-	113
1234A x E-126	1440/97	F <sub>4</sub>	100	46.29	97
1234B x E-112	1473/97	F <sub>4</sub>	100	40.80	99
Peredovik x E-112	1746/97	F <sub>4</sub>	75	-	97
1234B x E-112	1609/97	F <sub>4</sub>	100	42.43	114
E-112 x HA-821	1501/97	F <sub>4</sub>	100	41.01	114

Most interesting were the results showing the resistance of hybrid progenies to more than one pathogen. Some of these results are presented in Table 4. Different authors (Pustovoit and Gubin, 1974; Pustovoit and Krasnokutskaya, 1976) claim that wild species could be used in sunflower breeding for group immunity. Our results proved that interspecific hybrid material could be produced, carrying resist-

ance to two different pathogens. For example plants from four different cross combinations were resistant to both *Plasmopara helianthi* and *Orobanche cumana*, one progeny showed resistance to *Orobanche cumana* and *Phomopsis helianthi* and one to *Plasmopara helianthi* and *Phomopsis helianthi*. In most cases one of the resistances was less than 100%, which could be increased according to us by self pollination and accurate selection. As a conclusion we could say that interspecific hybridization has been and could be further successfully used for sunflower breeding for resistance to diseases and parasites.

Table 4: Evaluation of the reaction of hybrid material to more than one pathogen

Origin	Selection No.	Generation	Resistance to		
			<i>Pl.helianthi</i>	<i>O.cumana</i>	<i>Ph.helianthi</i>
Peredovik x E-126	668/97	F <sub>2</sub>	0	25	50
1234A x E-126	1440/97	F <sub>4</sub>	100	0	100
1234A x E-112	1058/97	F <sub>3</sub>	15.0	10	-
HA-300A x E-112	1063/97	F <sub>3</sub>	100	70	-
E-112 x HA-821	1068/97	F <sub>3</sub>	23.6	100	-
2607A x E-126	1175/97	F <sub>3</sub>	100	11.1	-

## CONCLUSIONS

Interspecific hybrid material has been produced by crossing cultivated sunflower inbred lines and opv. Peredovik with the wild annual *Helianthus annuus* that had showed resistance to *Plasmopara helianthi*, *Orobanche cumana* or *Phomopsis helianthi*. Some progenies were resistant to two different pathogens simultaneously. There were cases when self-pollination helped to increase the percentage of resistance up to 100%. Accessions GT-E-112 and GT-E-126 of the wild species *Helianthus annuus* could be successfully used as donors for resistance to *Plasmopara helianthi*, *Phomopsis helianthi* and *Orobanche cumana*.

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## RESISTENCIA A LAS ENFERMEDADES OBTENIDA POR LA HIBRIDIZACION INTERSPECIES

### RESUMEN

Para dos números de la especie silvestre del girasol *Helianthus annuus* L., GT-E-112 y GT-E-126, fue constatado que poseían los genes de resistencia a *Plasmopara helianthi* Novot., *Phomopsis helianthi* Munt.-Cvet. et al. y *Orobanche cumana* Wallr. El material creado por la hibridación interspecies con el girasol cultivado sensible mostró la resistencia a todos tres patógenos. Algunos descendientes poseían la resistencia simultánea a dos patógenos. Por la autofecundación, el porcentaje de resistencia se aumentó a 100%. Los números GT-E-112 y GT-E-126 pertenecen a la especie silvestre del girasol *Helianthus annuus* pueden ser utilizados con suceso como donadores de la resistencia a los patógenos *Plasmopara helianthi*, *Phomopsis helianthi* y *Orobanche cumana*.

## RÉSISTANCE À LA MALADIE OBTENUE PAR HYBRIDATION INTERSPECIES

### RÉSUMÉ

Il a été établi que deux numéros de l'espèce sauvage de tournesol *Helianthus annuus* L., GT-E-112 et GT-E-126 possèdent un gène de résistance aux *Plasmopara helianthi* Novot., *Phomopsis helianthi* Munt.-Cvet. et al. et à l'*Orobanche cumana* Wallr. Le matériel créé par hybridation interspecies avec le tournesol de culture, plus sensible, a montré une résistance aux trois pathogènes. Quelques lignées possédaient une résistance simultanée aux trois pathogènes. Quelques descendants possédaient une résistance simultanée à deux pathogènes. Le pourcentage de résistance a été augmenté à 100% par autofécondation. Les numéros GT-E-112 et GT-E-126 qui appartiennent à l'espèce sauvage de tournesol *Helianthus annuus* peuvent être utilisés avec succès comme donateurs de résistance aux pathogènes *Plasmopara helianthi*, *Phomopsis helianthi* et *Orobanche cumana*.

