

ACTIVE COLLECTION OF SUNFLOWER WILD HELIANTHUS SPECIES FROM NARDI FUNDULEA AND THEIR USE FOR THE INTROGRESSION OF RESISTANCE GENES TO THE RACES OF BROOMRAPE PRESENT IN ROMANIA IN CULTIVATED SUNFLOWER

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Abstract

Annual wild and perennial sunflower species are used in sunflower breeding programs for resistance to various biotic and abiotic factors. For increase the variability inside sunflower must use this species who represent a valuable gene pool. The parasite broomrape (*Orobanche cumana* Wallr) is a danger to the sunflower culture in Romania, especially in the southeastern part of the country where the most virulent races are present. The identification of new sources of resistance to the attack of this parasite is a priority in the sunflower breeding program from NARDI Fundulea. We have in our collection at NARDI Fundulea, mine perennial wild species and six annual wild species of sunflower and we cross gene from this species in elite line for improving resistance at parasite broomrape and other diseases of sunflower. By crossing the wild sunflower species with different cultivated sunflower genotypes, we obtained interspecific hybrids that are tolerant to more than race G and H of broomrape from area Braila in year 2017. These interspecific hybrids are sources of resistance to the races of broomrape present in Romania and can be used in breeding programs to obtain commercial hybrids of sunflower.

Key words: sunflower, broomrape, wild species, resistance, testing

In romanian agriculture, sunflower (*Helianthus annuus* L.) is principal oilseed crop and for that we must obtain sunflower hybrids with high oil content (more than 52%), resistant to principal diseases: downy mildew (*Plasmopara halstedii*), sunflower stalk and head rot (*Sclerotinia sclerotiorum*), sunflower rust (*Puccinia helianthi* Schwein), Phomopsis brown stem canker (*Diaporthe helianthi/Phomopsis helianthi* Munt), Verticillium wilt (*Verticillium dahliae* Kleb.), Phoma black stem (*Phoma macdonaldii* Boerma), Alternaria leaf spot (*Alternaria helianthi* Hansf.), Rhizopus head rot (*Rhizopus spp.*), Powdery mildew (*Erysiphe cichoracearum* DC. Ex Meret), Charcoal rot (*Macrophomina phaseolina* Tassi Goid) who limitate seed yield. For sunflower breeding, wild species of genus *Helianthus* (12 annual and 39 perennial) represent a very important resources for resistance to many pathogens and parasite broomrape *Orobanche cumana* Wallr. (Christov M., 2013; Jan C. *et al*,

2014; Seiler G. *et al*, 2017).

The most virulent regions from Romania infested with broomrape is Braila, Tulcea, and Constanta with races more than G. (Păcureanu Joița M., 2014; Rîșnoveanu L. *et al*, 2016; Anton F.G. *et al*, 2016).

For the broomrape parasite (*Orobanche cumana* Wallr), resistance genes have been identified in wild sunflower species. Wild annual species poses resistance at broomrape at race E: *Helianthus annuus* and *Helianthus debilis* ssp. *Debilis*; at race E-F: *Helianthus deserticola* and *Helianthus exilis*; at races E-H: *Helianthus argophyllus* and *Helianthus petiolaris*. (Christov M. *et al*, 2009; Antonova T.S. *et al*, 2011; Terzic S. *et al*, 2010, 2016; Seiler G., 2018).

Inbred line derived from *Helianthus deserticola* and *Helianthus anomalus* was resistant to race E to broomrape (Hladni N. *et al*, 2009, 2010; Fernandez-Martinez J., 2000). Gene Or_{deb} was identified in Spain in inbred line derived from

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H. debilis subsp. *Tardiflorus* who have resistance at race G of broomrape (Velasco L. *et al*, 2012). Gene O_{pral} was identifying in annual wild specie *Helianthus praecox* who give resistance at race G (Sayago A. *et al*, 2018).

From interspecific hybridization between perennial species with sunflower cultivated was obtained inbred line resistance at race E-F: *Helianthus maximiliani*, *Helianthus nuttallii*,

Helianthus pauciflorus and *Helianthus resinosus*; at race F: *Helianthus hirsutus*, *Helianthus grosseserratus*; at race F-G: *Helianthus divaricatus*, *Helianthus eggertii*, *Helianthus smithii* and *Helianthus tuberosus* (Jan C. *et al*, 2000; Christov M., 2013; Anton F.G. *et al*, 2017).

In our active collection we have 9 wild perennial species with 31 accesions and 6 wild annual species with 24 accesions (table 1).

Table 1

Active collection with annual and perennial wild species of genus *Helianthus* from NARDI Fundulea

No. field	Annual/ Perennial	Species	Origin	Height	Head diameter	Flowering time
P 1 M	Perennial	<i>Helianthus maximiliani</i>	Bulgaria 2014 GT-M-017	250 cm	3 cm	End of September
P 2 M	Perennial	<i>Helianthus maximiliani</i>	Bulgaria 2014 GT-M-017A	300 cm	3 cm	Beginning of October
P3 M	Perennial	<i>Helianthus maximiliani</i>	Bulgaria 2014 GT-M-177	210 cm	1.5 cm	Middle of June
P4 M	Perennial	<i>Helianthus maximiliani</i>	Bulgaria 2014 GT-M-018	240 cm	1.7 cm	Beginning of October
P 5 M	Perennial	<i>Helianthus maximiliani</i>	Bulgaria 2014 GT-M-175	185 cm	2.5 cm	Beginning of August
P 6 H	Perennial	<i>Helianthus hirsutus</i>	Bulgaria 2014 GT-M-164	250 cm	1.7 cm	Middle of July
P 7 D	Perennial	<i>Helianthus decapetalus</i>	Bulgaria 2014 GT-M-171	190 cm	1.7 cm	Middle of July
P 8 St	Perennial	<i>Helianthus strumosus</i>	Bulgaria 2014 GT-M-110	250 cm	2 cm	Beginning of August
P 9 Mo	Perennial	<i>Helianthus mollis</i>	Bulgaria 2014 GT-M-082A	165 cm	2.5 cm	Beginning of July
P 10 St	Perennial	<i>Helianthus strumosus</i>	Bulgaria 2014 GT-M-059	225 cm	2 cm	End of July
P 11 T	Perennial	<i>Helianthus tuberosus</i>	Bulgaria 2014 GT-M-146	240 cm	1 cm	End of August
P 12 Di	Perennial	<i>Helianthus divaricatus</i>	Bulgaria 2014 GT-M-129	160 cm	1.5 cm	Beginning of July
P 13 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-13	270 cm	1.7 cm	End of July
P 14 M	Perennial	<i>Helianthus maximiliani</i>	Romania 2016 FD-M-14	180 cm	1 cm	End of June
P 15 G	Perennial	<i>Helianthus giganteus</i>	Bulgaria 2014 GT-M-153	265 cm	2 cm	Beginning of August
P 16 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-16	240 cm	2 cm	Beginning of September
P 17 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-17	215 cm	2 cm	End of July
P 18 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-18	290 cm	2 cm	End of July
P 19 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-19	270 cm	2 cm	End of July
P 20 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-20	275 cm	2 cm	End of July
P 21 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-21	275 cm	2 cm	End of July
P 22 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-22	260 cm	2 cm	End of July
P 23 T	Perennial	<i>Helianthus tuberosus</i>	Romania 2016 FD-T-23	260 cm	2 cm	End of July
P 24 G	Perennial	<i>Helianthus giganteus</i>	Serbia 2016 GIG 2017	175 cm	1.5 cm	Middle of August
P 25 Gro	Perennial	<i>Helianthus groseratus</i>	Serbia 2016 GRO 2022	190 cm	0.7 cm	Beginning of August
P 26 Gro	Perennial	<i>Helianthus groseratus</i>	Serbia 2016 GRO 2039	195 cm	1.5 cm	End of July
P 27 T	Perennial	<i>Helianthus</i>	Serbia 2016	120 cm	3 cm	End of July

		<i>tuberosus</i>	TUB 15			
P 28 T	Perennial	<i>Helianthus tuberosus</i>	Serbia 2016 TUB 1704	140 cm	1 cm	End of July
P 30 M	Perennial	<i>Helianthus maximiliani</i>	Serbia 2016 MAX 041	120 cm	1 cm	Middle of June
P 31 M	Perennial	<i>Helianthus maximiliani</i>	Serbia 2016 MAX 2195	65 cm	0.5 cm	End of June
P 32 M	Perennial	<i>Helianthus maximiliani</i>	Serbia 2016 MAX 2227	140 cm	0.7 cm	Middle of June
No. field	Annual/Perennial	Species	Origin	Height	Head diameter	
A 1 Ne	Annual	<i>Helianthus neglectus</i>	Bulgaria 2014 017	115 cm	2.5 cm	
A 2 De	Annual	<i>Helianthus debilis</i>	Bulgaria 2014 014	140 cm	2 cm	
A 3 De	Annual	<i>Helianthus debilis</i>	Bulgaria 2014 136	150 cm	2.5 cm	
A 4 De	Annual	<i>Helianthus debilis</i>	Bulgaria 2014 137	120 cm	2 cm	
A5 Pe	Annual	<i>Helianthus petiolaris</i>	Bulgaria 2014 020	80 cm	2 cm	
A6 Pe	Annual	<i>Helianthus petiolaris</i>	Bulgaria 2014 022	130 cm	1.5 cm	
A7 Pe	Annual	<i>Helianthus petiolaris</i>	Bulgaria 2014 023	125 cm	2 cm	
A8 Pe	Annual	<i>Helianthus petiolaris</i>	Bulgaria 2014 142	130 cm	2.5 cm	
A9 Pr	Annual	<i>Helianthus praecox</i>	Bulgaria 2014 145	125 cm	2 cm	
A10 Pr	Annual	<i>Helianthus praecox</i>	Bulgaria 2014 148	120 cm	2 cm	
A11 Pr	Annual	<i>Helianthus praecox</i>	Bulgaria 2014 150	135 cm	2 cm	
A12 Pr	Annual	<i>Helianthus praecox</i>	Bulgaria 2014 151	120 cm	2.5 cm	
A13 Arg	Annual	<i>Helianthus argophyllus</i>	Romania 2014 FD-ARG-13	140 cm	2 cm	
A14 An	Annual	<i>Helianthus annuus</i>	Serbia 2016 A 03	160 cm	3 cm	
A16 An	Annual	<i>Helianthus annuus</i>	Serbia 2016 ANN 0340	195 cm	4cm	
A20 An	Annual	<i>Helianthus annuus</i>	Serbia 2016 ANN 2201	75 cm	1.5 cm	
A21 Arg	Annual	<i>Helianthus argophyllus</i>	Serbia 2016 ARG 1317	165 cm	3 cm	
A23Arg	Annual	<i>Helianthus argophyllus</i>	Serbia 2016 ARG 1677	200 cm	3.3 cm	
A24 Arg	Annual	<i>Helianthus argophyllus</i>	Serbia 2016 ARG 1805	185 cm	3 cm	

MATERIAL AND METHOD

We collected broomrape seeds from Braila area in year 2017 and we tested in year 2018, in artificial condition, in greenhouse in Fundulea, resistance at broomrape of annual species of genus *Helianthus* and of sunflower interspecific populations. We used 10 buckets of 10 l on every species/genotype tested with 10 seed of sunflower per bucket. We put mixed fertile soil and sand, and 2 g of broomrape seed/bucket. After 35 days and 47 from sowing we make observation about infestation with broomrape on root of sunflower.

RESULTS AND DISCUSSIONS

Wild species have a low germination and because of that is very difficult to obtain many mature plant. We tested only annual species for resistance to broomrape with the best germination.

Eight accession of three annual species don't have broomrape attached on roots and other eleven accessions of five annual species are susceptible to races of broomrape present in area Braila 2017 (table 2).

Table 2

Resistance of annual <i>Helianthus</i> species at broomrape infestation in year 2018, in greenhouse		Number of broomrape/sunflower plant after 35 days from sowing (population of broomrape from Braila 2017 area)	Number of broomrape/sunflower plant after 47 days from sowing (population of broomrape from Braila 2017 area)
A1Ne	<i>Helianthus neglectus</i>	0 broomrape/14 plants	12 broomrape/12 plants
A2De	<i>Helianthus debilis</i>	0 broomrape/18 plants	13 broomrape/13 plants
A3De	<i>Helianthus debilis</i>	0 broomrape/7 plants	10 broomrape/10 plants
A4De	<i>Helianthus debilis</i>	0 broomrape/4 plants	0 broomrape/7 plants
A5Pe	<i>Helianthus petiolaris</i>	0 broomrape/10 plants	0 broomrape/10 plants
A6Pe	<i>Helianthus petiolaris</i>	0 broomrape/12 plants	0 broomrape/8 plants
A7Pe	<i>Helianthus petiolaris</i>	0 broomrape/30 plants	0 broomrape/7 plants
A8Pe	<i>Helianthus petiolaris</i>	0 broomrape/16 plants	0 broomrape/6plants
A9Pr	<i>Helianthus praecox</i>	0 broomrape/11 plants	0 broomrape/5 plants
A10Pr	<i>Helianthus praecox</i>	0 broomrape/12 plants	0 broomrape/7 plants
A11Pr	<i>Helianthus praecox</i>	0 broomrape/12 plants	0 broomrape/3 plants
A12Pr	<i>Helianthus praecox</i>	0 broomrape/7 plants	9 broomrape/9 plants
A13Arg	<i>Helianthus argophyllus</i>	5 broomrape/3 plants	25 broomrape/5 plants
A14 An	<i>Helianthus annuus</i>	0 broomrape/5 plants	6 broomrape/6 plants
A16 An	<i>Helianthus annuus</i>	5 broomrape/5 plants	90 broomrape/3 plants
A20 An	<i>Helianthus annuus</i>	2 broomrape/2 plants	14 broomrape/2 plants
A21 Arg	<i>Helianthus argophyllus</i>	13 broomrape/4 plants	16 broomrape/4 plants
A23 Arg	<i>Helianthus argophyllus</i>	46 broomrape/13 plants	12 broomrape/4 plants
A24 Arg	<i>Helianthus argophyllus</i>	55 broomrape/11 plants	15 broomrape/5 plants

In table 3 we present some agronomic characteristics about 20 interspecific hybrids which we tested for resistance to broomrape.

Head diameter of this 20 interspecific hybrids is between 10 cm and 28 cm, height is

between 80 cm and 175 cm, seed yield per plant is between 6.19g and 51.26g, days from sowing to maturity is between 110 and 150.

Table 3

Some agronomic characteristic of interspecific hybrids tested

No	Interspecific Population	Head diameter	Height	Seed yield/plant	Branching\ Non branching	Days to maturity
1	16471B x A1Ne	10 cm	80 cm	29.00g	Branching	110
2	602B x A1Ne	12cm	110 cm	8.71g	Branching	120
3	3249B x A1Ne	18 cm	130 cm	11.53g	Non branching	140
4	1010B x A2De	20 cm	172 cm	36.52g	Non branching	150
5	1029B x A2De	22 cm	123 cm	6.83g	Non branching	150
6	0925C x A2 De	21 cm	119 cm	18.82g	Non branching	140
7	24 B x A3 De	28 cm	135 cm	6.19g	Non branching	150
8	885 BxA3De	13 cm.	132 cm	11.03g	Branching anthocyanin coloration	125
9	7869B x A8Pe	23 cm	125 cm	28.06g	Non branching	140
10	602B x A9Pr	11 cm	100 cm	20.54g	Branching	120
11	19669B x A10Pr	13 cm	120 cm	7.81g	Branching	100
12	16438B x A13Arg	19 cm	94 cm	24.90g	Non branching	150
13	1068C x P14M	11 cm	130 cm	12.69g	Branching	120
14	16438B x P11T	21 cm	95 cm	10.07g	Non branching	150
15	16471B x P15G	16 cm	85 cm	33.60g	Branching	120
16	1633B x P12D	18 cm	96 cm	26.26g	Non branching	150
17	3240B x P3M	17 cm	125 cm	25.31g	Non branching	150
18	1064C x P9Mo	24 cm	116 cm	10.18g	One lateral branching	140
19	1001B x P6H	23 cm	146 cm	11.34g	One lateral branching	140
20	1050B x P8St	24 cm	175 cm	51.26g	Non branching	140

We tested 20 interspecific hybrids in artificial condition and after 47 days after sowing we make notation about attack degree of

broomrape and intensity of broomrape attack (table 4).

Table 4

Resistance of interspecific hybrids at broomrape infestation from Braila 2017 area, in greenhouse, in year 2018

No./2018	Interspecific Population	Attack degree of broomrape (%)	Intensity of broomrape attack
1	16471B x A1Ne	49%	16.1
2	602B x A1Ne	32%	16.2
3	3249B x A1Ne	35%	13.1
4	1010B x A2De	53%	15.8
5	1029B x A2De	48%	5.1
6	0925C x A2 De	44%	13.6
7	24 B x A3 De	45%	15.5
8	885 BxA3De	46%	15.5
9	7869B x A8Pe	35%	4.8
10	602B x A9Pr	37%	3.2
11	19669B x A10Pr	38%	5.1
12	16438B xA13Arg	30%	24.1
13	1068C x P14M	16%	6.4
14	16438B x P11T	52%	8
15	16471B x P15G	45%	11.6
16	1633B x P12D	52 %	11.2
17	3240B x P3M	47%	7.4
18	1064C x P9Mo	41%	11
19	1001B x P6H	47%	7.8
20	1050B x P8St	51%	6.6

CONCLUSIONS

It is very important to tested in every year new genotype of sunflower and many wild species of genus *Helianthus* to identified resistant/tolerant sources for broomrape. Some accessions of *Helianthus debilis*, *Helianthus petiolaris* and *Helianthus praecox* have total resistance at races of broomrape present in Braila 2017 area and interspecific hybrids with this species have a good tolerance. Interspecific hybrid 1068C x P14 M result from crossing with *Helianthus maximiliani* have a better tolerance then other interspecific hybrids.

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